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Society of Environmental Toxicology and Chemistry

ABSTRACT BOOK

SETAC

SETAC 19TH ANNUAL MEETING

The Natural Connection: Environmental Integrity and Human Health

15 - 19 November 1998
Charlotte, NC

ABSTRACT BOOK

SETAC 19TH ANNUAL MEETING

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This book is comprised of the abstracts of the presentations for the platform, poster, and interactive poster sessions of the SETAC 19th Annual Meeting of the Society of Environmental Toxicology and Chemistry (SETAC), held at the Charlotte Convention Center, Charlotte, NC, 15 - 19 November 1998.

The abstracts are reproduced as accepted by the Program Committee of the 19th Annual Meeting and appear in numerical order.

This book contains an Author Index that cross-references the corresponding abstract number(s). A Key Word Index (by subject) to all the presentations is also included.

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In the 1970's, no forum existed for interdisciplinary communication among environmental scientists -- biologists, chemists, toxicologists -- and others interested in environmental issues such as managers and engineers. The Society of Environmental Toxicology and Chemistry (SETAC) was founded in 1979 to fill the void. Based on the growth in membership, annual meeting attendance, and publications, the forum was needed.

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FACTS & FIGURES

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Sep 1998 / CM

001 Comparative Nutritional Ecology and Ichthyotoxicity of Two Morphospecies Within the "Toxic *Pfiesteria* Complex". Burkholder, J. M. and Howard Glasgow, North Carolina State University, Raleigh North Carolina; Lewitus, A. J.; University of South Carolina, Georgetown , South Carolina. We compared the response to N and P enrichments, and toxicity toward fish of the toxic dinoflagellate, *Pfiesteria piscicida* and a second *Pfiesteria*-like species that were isolated (unispecies but not axenic) from two known fish kill sites in the Neuse Estuary, NC. Both dinoflagellates were acclimated in separate cultures for two months under similar conditions in which they were (a) grown with fish or (b) grown with a cryptomonad as algal prey. Nontoxic zoospore production (i.e., in the absence of fish) was then compared for each species in separate trials across gradients of nitrate, ammonium, inorganic phosphate, and organic phosphate enrichment in short-term batch cultures, with vs. without an N/P-starved cryptomonad as available algal prey (sufficient prey added to achieve an initial concentration of 5,000 or 50,000 cells/ mL). The two species differed significantly in response to both fish and nutrients/algal prey, in timing and overall cell production. *Pfiesteria piscicida* responded more slowly and attained significantly lower cell densities when given algae, especially following a diet of live fish. Highest growth of both dinoflagellates occurred with nitrate stimulation and tracked growth of the cryptomonad prey. Our isolate of *Pfiesteria piscicida* was tested as significantly more toxic to fish than the *Pfiesteria*-like species, with 100-fold higher cell density or much longer periods required for lethality from the *Pfiesteria*-like species. Thus far the data indicate that the second known *Pfiesteria*-like species isolated from natural habitat is only weakly toxic to fish, relative to *Pfiesteria piscicida.*

002 *Pfiesteria piscicida:* Development of a DNA Probe. Rublee, Parke. A., Kristen L. Toffer, Eric F. Schaefer, and Jason W. Kempton, Biology Department, University of North Carolina at Greensboro, Greensboro, NC 27412; JoAnn M. Burkholder, and Howard B. Glasgow, Jr.; Botany Dept., North Carolina State University, Raleigh, NC 27695. To facilitate rapid identification of the toxic ambush-predator Pfiesteria piscicida, we have developed extraction protocols, sequenced SSU rDNA from a Pfiesteria piscicida culture from the Pamlico River, NC, and located unique regions of the SSU rDNA as targets for molecular probes. Several pairs of primers have effectively amplified DNA from cultures of Pfiesteria piscicida isolated from different geographic areas along the eastern United States, but not from other dinoflagellates. Flourescent oligonucleotide probes designed from the primers also distinguish Pfiesteria piscicida from other dinoflagellates. We are refining our methods for field assays to detect Pfiesteria at sub-lethal concentrations in estuarine waters beginning in the summer of 1998.

003 Reporter Gene Assay for Fish Killing Activity Produced by *Pfiesteria piscicida.* J. S. Ramsdell, E.R. Fairey, J. S. Edmunds, N. J. Deamer-Melia, H. Glasgow Jr., F. M. Johnson, P. R. Moeller and J. M. Burkholder. National Ocean Service Center for Coastal Environmental Health and Biomolecular Research, Charleston, SC and North Carolina State University, Raleigh, NC. Collaborative studies were held to develop a functional assay for fish- killing toxins produced by Pfiesteria piscicida. Eight cells lines were used to screen organic fractions by using a MTT cytotoxicity assay. Diethyl ether and residual water after extraction were cytotoxic to several cell lines including GH4C1 rat pituitary cells. Residual water as well a preextracted culture water containing Pfiesteria piscicida cells induced c-fos-luciferase expressed in GH4C1 cells with a rapid time course of induction and sensitive detection. The reporter gene assay detected activity in toxic isolates of P. piscicida from two North Carolina estuaries and may be suitable for detecting toxic activity in human and animal serum.

004 Identification and Differentiation of Morphospecies in the Toxic *Pfiesteria* Complex. Glasgow,H.B.; Burkholder,J.M.; Hannon, E.K.; North Carolina State University, Raleigh, North Carolina. Recent toxic outbreaks by the dinoflagellate, *Pfiesteria piscicida*, have provided a compelling illustration of strong linkages between fish kills/epizootics and subtle but serious impacts on human health. *P. piscicida* co-occurs with at least one other *Pfiesteria*-like species (not yet named) that has tested as more weakly toxic; this species typically is outcompeted by *P. piscicida* in active fish-killing cultures. Thus, there are two known species in the "toxic *Pfiesteria* complex" thus far, both found to date in the Albemarle-Pamlico and the Chesapeake. These two species cannot be discerned using light microscopy (LM) but can be clearly differentiated under SEM following osmotic procedures to well their plate sutures. In addition to specific morphological characteristics true *Pfiesteria*-like species must possess the following characteristics: (i) toxicity toward fish; (ii) attack behavior toward live fish; (iii) lacking photosynthesis, or, alternatively, having photosynthesis from kleptochloroplasts located in an epithecal food vacuole; and (iv) complex life cycle with amoeboid stages. Current methods for culturing toxic and nontoxic stages of the *Pfiesteria* species "complex:" include(i) bioassays with live fish in biohazard III facilities (toxic stages) and (ii) using various algal prey (nontoxic). We compared zoospore production for *Pfiesteria piscicida* and a 2nd toxic *Pfiesteria*-like species using these two culture methods. Field samples from Chesapeake Bay and the Albemarle-Pamlico were subaliquoted, and subsamples were cultured with fish or with cryptomonad prey over a 8-week time series. At least 100 cells prepared with suture-swelling procedures were evaluated to determine thacal plate tabulation for morphospecies ident!!ification (ca. 2,500 cells screened over 2.5 days per replicate). In 7 of 8 comparisons, the fish bioassay yielded only *Pfiesteria piscicida*; in 1 case, a mix of *P. piscicida* (60%), the 2nd toxic *Pfiesteria*-like species (10%), and *Gyrodinium galetheanum* (20%) occurred. By contrast, algal bioassays consistently yielded species mixes that were dominated by various nontoxic species or by the 2nd potentially toxic *Pfiesteria*-like species, with *P. piscicida* less common or not detected. The data indicate that (i) the algal assay is less specific for detecting *P. piscicida* than fish bioassays; and (ii) under the culture conditions used, *P. piscicida* outcompetes other algae in the presence of live fish, whereas the 2nd potentially toxic *Pfiesteria*-like species and other mixotrophic dinoflagellates outcompete *P. piscicida* for algal prey.

005 Aspects of the Nutritional Physiology of *Pfiesteria piscicida.* Lewitus, Alan J., JoAnn M. Burkholder, Howard B. Glasgow Jr. and Patricia M. Glibert. University of South Carolina (AJL), Baruch Marine Laboratory, P.O. Box 1630, Georgetown, SC 29442, North Carolina State University (JMB, HBG), Botany Department, P.O. Box 7612, Raleigh, NC 27695, and University of Maryland (PMG), Horn Point Laboratory, P.O. Box 775, Cambridge, MD 21613. Like many dinoflagellates, Pfiesteria piscicida is characterized by complex nutritional physiological responses; i.e. its growth can be supported by diverse substrate forms and by various modes of nutrition. This nutritional versatility complicates the identification of potential links between nutrient enrichment and P. piscicida proliferation and toxic activity. Although nontoxic zoospores of P. piscicida have a relatively proficient grazing ability, they also can function as phototrophs by acquiring functional chloroplasts from phytoplankton prey, a process termed kleptoplastidy. The nutritional responses of P. piscicida to inorganic and organic nutrient additions were compared in heterotrophic zoospores feeding on cryptophytes vs. phototrophic cultures with cryptophyte kleptochloroplasts. Nutrient additions stimulated the growth of heterotrophic cultures indirectly (by increasing the abundance of cryptophyte prey), although direct uptake of 15N-labeled organic and inorganic substrates was detected. Phototrophic cultures also were capable of taking up both classes of nutrients, with uptake rates of some substrates, such as urea, exceeding those of heterotrophic cultures. The results suggest that knowledge of the types of nutrients involved in stimulating P. piscicida growth, and the pathways of nutrient acquisition, depends on understanding the ecophysiology of P. piscicida's nutritional state (heterotrophy vs. phototrophy vs. mixotrophy).

006 Behavioral and Physiological Responses of Three Commercially Important Species of Bivalves to the Toxic Estuarine Dinoflagellate, *Pfiesteria piscicida*. Springer 1, J.J., Cancillieri 2, P., Shumway 2, S.E., Glasgow 1, H.B., and Burkholder 1, J.M. . 1 North Carolina State University, Dept. of Botany, Box 7612, Gardner Hall, Raleigh, NC 27695-7612. 2 Southampton College of Long Island University, Natural Sciences Division, Southampton, NY 11968. The toxic estuarine dinoflagellate, *Pfiesteria piscicida*, has been implicated as the causative agent in major fish kills along the eastern seaboard of the U.S. *P. piscicida*, when stimulated by the presence of finfish, produces toxin(s) which strip epidermal tissue from finfish and impair the nervous system leading to paralysis and suffocation. The responses of Argopecten irradians (northern bay scallop), Crassostrea virginica (eastern oyster), and Mercenaria mercenaria (northern hardclam), to P. piscicida and its associated toxin(s) were examined. The extent to which these species accumulate P. piscicida's toxin(s) and the range of chronic/sublethal impacts on major life history stages including larvae, spat, juveniles, and adults were documented.

007 A Rat Model of the Neurocognitive Effects of Exposure to *Pfiesteria piscicida*. ED Levin[1], B Simon[1], D Schmechel[1], H Glasgow, Jr.[2], N Deamer-Melia[2], JM Burkholder[2]. [1]Integrated Toxicology Program, Duke University, Durham, NC, [2]Department of Botany, North Carolina State University, Raleigh, NC. *Pfiesteria piscicida* is an estuarine dinoflagellate responsible for mass fish kills in North Carolina and other areas along the eastern coast of the United States. We have developed a rat model of *Pfiesteria* effects on neurocognitive function. In four different studies, including 36 controls and 48 *Pfiesteria*-treated female Sprague-Dawley rats, a single sc injection of 106,800-320,400 cells/kg of *Pfiesteria* caused a persistent learning deficit ($p<0.005$) in the radial-arm maze . The deficit was also seen in rats administered a filtered cell-free preparation equivalent to 106,800 cells/kg. The learning deficit did not seem to result from motivational, sensorimotor or memory deficits. Rats pretrained on the maze prior to *Pfiesteria* administration did not show deficits, but they did show deficits in learning a new task ten weeks after administration. A lower dose of 35,600 *Pfiesteria* cells/kg was not effective in producing a deficit. The *Pfiesteria*-induced learning deficits were seen in rats that did not otherwise show health impairments. The learning deficits caused by *Pfiesteria* treatment in these studies may be related to cognitive deficits seen in humans after accidental *Pfiesteria* exposure.

008 Human Health Effects Associated with Laboratory Exposures to *Pfiesteria piscicida*. Schmechel, D.E., Koltai, D.C., Welsh-Bohmer, K.A.. Neurological Disorders Clinic, Departments of Medicine and Psychiatry, Duke University Medical Center, Durham, NC 27705. Five patients with laboratory exposures to cultures of the estuarine dinoflagellate *Pfiesteria piscicida* have undergone comprehensive clinical evaluation at our clinic. Mechanism of, degree, and time of exposure varied across patients. However, the most severe case had both contact and aerosol exposure to *Pfiesteria*. Complaints of the patients included subacute physical and/or neurocognitive symptoms. Neuroimaging studies including MRI and PET scans were generally within normal limits. Relative weaknesses in learning and memory skills and complex information processing abilities were observed for some individuals on neuropsychological evaluation. Longitudinal follow-up of some cases indicates the likelihood of resolution of symptomatology with exposure cessation. Most persons reportedly resolve severe symptoms in a few hours to days after cessation of exposure. The slow resolution of other significant neurocognitive symptoms and findings (weeks to months) suggests that either the toxin may be stored in body tissues (e.g., lipophilic) and/or the effects on neural systems are sustained by some other mechanism. It is clear that significant variation in clinical presentation occurs which may reflect amount of toxin, route of exposure, premorbid patient characteristics, and/or possible *Pfiesteria* subtypes or multiple toxins. Evaluation of cases referred to our clinic with putative environmental exposures (approximately seven cases from NC and MD) support this observed clinical variability. Medical evaluation of these individuals indicate potential human health effects from exposure to *Pfiesteria* environmentally. However, alternative diagnostic considerations are frequent, and thus require close attention in this group. The laboratory exposure cases offer the clearest observations of the health effects associated with exposure to high levels of this dinoflagellate and/or its toxins.

009 Epidemiological Assessment of Pfiesteria Exposure in Humans. Morris, JG, Jr., Grattan, LM, Oldach D, Perl TM, Lowitt MH, Matuszak DL, Charache P, and *Pfiesteria* Research Group, University of Maryland and Johns Hopkins University Schools of Medicine, and Maryland Department of Health and Mental Hygiene, Baltimore, MD. Beginning in the fall of 1996, commercial fishermen "watermen" on the eastern shore of the Chesapeake Bay in Maryland began to note unusual "punched out" lesions on fish, and to have complaints of fatigue, respiratory irritation, and "memory problems." Fish lesions were associated with the presence of toxic *Pfiesteria* or *Pfiesteria*-like dinoflagellates in the Pocomoke River and near-by waterways. In neuropsychologic testing, persons with high levels of exposure to affected waterways had significant decreases in performance on tests measuring learning ability and higher order cognitive function. A dose-response effect was apparent, with degree of exposure correlating with degree of neuropsychologic deficit. The observed effects had largely resolved within 6-8 weeks of cessation of exposure; within 6 months, all persons tested had test scores which were within normal ranges. Risk of illness was increased in association with contact with fish with lesions; no association was seen with ingestion of seafood. Our data suggest that exposure to water containing toxic *Pfiesteria* or *Pfiesteria*-like dinoflagellates results in a reversible clinical syndrome characterized by difficulties with learning and memory.

010 Pfiesteria Symposium Summary and Review. Kenneth H. Reckhow, UNC, Water Resources Research Institute. Research results presented in the symposium are briefly summarized. In addition, research and management recommendations from a recent workshop in North Carolina are presented. Finally, ambient water quality monitoring results in the Neuse River during the summer of 1998 are examined.

011 Measuring Oxidation/Reduction Zones, Calculating Degradative Rates and Integration with A Groundwater Solute Transport Model: A Complicated Case Study of "Zones within Zones" for Natural Attenuation. McInnis, D. L., Toxicological & Environmental Associates, Inc., Baton Rouge, LA; Droy, B. F., Ph.D., Toxicological & Environmental Associates, Inc., Baton Rouge, LA; Pardue, J., Ph.D., Department of Civil and Environmental Engineering, Louisiana State University, Baton Rouge, LA. Groundwater geochemistry data, (volatile organic compounds, dissolved gases, hydrogen, nitrate, nitrite, sulfate, sulfide, ferrous iron, chloride, and others) from a Superfund Site in Louisiana, were collected and used to determine the zonations of existing oxidation/reduction processes occurring in the aquifers underlying the site. These data demonstrate alternating redox zones (zones within zones) encountered by the contaminated groundwater flowing throughout the aquifer as well as the presence of daughter products resulting from reductive dechlorination of chlorinated organic compounds. The results of the geochemistry data were then used to calculate degradative rate constants. These results were incorporated into the site solute fate and transport model to further assess the effectiveness of natural attenuation of the volatile organic contaminants. These results have demonstrated that chlorinated ethenes, in this system, are subject to sequential reductive and oxidation processes that favor natural attenuation. The sequential redox zone-specific results of reductive dechlorination of site constituents of concern will be presented.

012 Identification of Bacteria in Contaminated Ground Water Using the RNA-Hybridization Technique. Farmer, J.J., Byl, T.D., Tennessee State University and U.S. Geological Survey, Nashville, TN, USA; Williams, S.D., U.S. Geological Survey, Nashville, TN; and Bailey, F.C., Middle Tennessee State University and U.S. Geological Survey. The RNA-hybridization technique is a new method to identify microorganisms. The RNA hybridization technique can identify microorganisms by taking advantage of unique genetic sequences on ribosomal RNA. This technique was used to identify bacteria in karst ground water at a site contaminated with chlorinated solvents. Well samples of anaerobic ground water had a higher percentage of sulfate-reducing bacteria and methanogenic bacteria. These bacteria are known to cause anaerobic reductive dechlorination of chlorinated solvents. Well samples of ground water that fluctuated from anaerobic to aerobic contained sulfate-reducing bacteria, ammonia-oxidizing bacteria, and methane-oxidizing bacteria. The latter two are known to biodegrade chlorinated solvents through cometabolic processes. This bacterial information was used in conjunction with geochemical and hydrologic information to identify the most prominent pathways of biodegradation at the contaminated site.

013 Evaluating BOD, COD, and TOC as a Surrogate Measure of Food Replenishment for Bacteria in a TCE-Contaminated Karst Aquifer. Akula, S., Tennessee State University, Nashville, TN; Williams, S.D., and Byl, T.D., U.S. Geological Survey, Nashville, TN. Chemical and biological data collected at a TCE-contaminated karst site in Middle Tennessee indicate that biodegradation is taking place naturally at the site. If bioremediation is to be a corrective action at the site, food replenishment and sources for microorganisms should be evaluated. Biochemical oxygen demand (BOD), chemical oxygen demand (COD) and total organic carbon (TOC) were measured in samples from several wells at the site as possible indicators of food replenishment. Water-level data and additional water-quality data such as dissolved oxygen (DO), pH, specific conductance and temperature were monitored in-situ over a 2 month period to determine how hydrologic and water-quality changes may be related to food replenishment. Continuous field data show that DO in 100 foot-deep karst conduits was replenished within a few hours after rain events. Synoptic sampling has demonstrated that water in some wells was consistently low in DO (<0.1 mg/L) and high in COD regardless of weather conditions. Though BOD and TOC concentration in samples from these same wells appeared to vary in response to changing weather conditions, TOC did not appear to be well correlated with weather patterns. The correspondence of rain events with changes in BOD shows that BOD was generally highest under dry conditions indicating a possible lag time between food replenishment and BOD response. Lab experiments conducted on field samples and spiked with ammonia and acetate verified these findings. The addition of food in lab studies also stimulated the rate of biodegradation by 350 percent over that observed in sterile controls. Lower concentrations of food stimulated the biodegradation to a lesser extent.

014 Microcosm Study to Determine the Feasibility of Bioremediation at a KarstSite. Franklin, P.R.*, Tennessee State University, Nashville, TN;Williams, S.D., Farmer, J.J., and Byl, T.D., U.S. Geological Survey,Nashville, TN. The potential for bioremediation of trichloroethylene (TCE) is being examined at a karst site in Middle Tennessee where "pump and treat" wells are used to slow the movement of ground water away from the site and to remove some of the TCE. Ground-water samples were collected from nearby wells and analyzed for geochemical indicators of bioremediation (electron donors and acceptors). Trends in the concentrations of TCE and intermediate degradation products also were monitored. Bacteria associated with the degradation of TCE were identified in water samples using the RNA oligonucleotide hybridization technique. Water-quality data indicate that both aerobic and anaerobic conditions occur in the karst aquifer. Sulfate-reducing and methanogenic bacteria that are associated with the reductive dechlorination of TCE (an anaerobic process) were present in samples of anaerobic well water. Methanotrophs and ammonia-oxidizing bacteria, associated with co-metabolic degradation of TCE (an aerobic) process), were present in samples of aerobic well water. Aerobic and anaerobic bacteria were identified in well water that fluctuated in dissolved-oxygen concentrations. Microcosm studies of ground water collected from the karst aquifer indicated that both biodegradation pathways are active. The biodegradation in some well water was rapid, with a TCE half-life of 35 days. Additional water-quality and hydrologic monitoring of the karst aquifer are being conducted to determine temporal and spatial changes in water quality and to evaluate how these changes may affect the biodegradation processes at the site.

015 Phytoremediation Of Dissolved Phase Trichloroethylene Using Mature Vegetation. Doucette, W.J.*, Bugbee, B. and Hayhurst, S. C. Utah State University, Logan, UT. A field study was conducted to determine if existing, mature vegetation is involved in the uptake, volatilization and/or metabolism of trichloroethylene (TCE) from a shallow contaminated groundwater plume. Four on-site plants were sampled, three in the plume area and one outside. One additional off-site control plant was also sampled. Groundwater concentrations of TCE below the vegetation ranged from 0.4 to 90 mg/L. Flow-through glass chambers were used to collect transpiration gases for TCE analysis. Leaf, stem and root samples were collected and analyzed for TCE and 3 metabolites (2,2,2-trichloroethanol [TCEt], 2,2,2-trichloroacetic acid [TCAA] and 2,2'-dichloroacetic acid [DCAA]) identified in previous laboratory studies. Measurable levels of TCE were found in 7 of 15 transpiration gas samples and 2 of 3 apparatus blanks. TCE (6 of 34 samples), TCEt (10 of 31), TCAA (22 of 27), or DCAA (3 of 27) were identified in all four plants from the site. The off-site control plant showed no measurable levels of TCE or its metabolites, suggesting that the plant thought to be outside of the plume area had been exposed to TCE. Yearly precipitation patterns and root distribution (70% in top 2 ft, 90% in top 4 ft) imply that direct groundwater use by existing vegetation is much less than that obtained by precipitation. This may reduce the impact of vegetation on the removal of TCE at this site. Similar measurements at more arid sites, where direct groundwater use by vegetation is typically greater, must be made before conclusions regarding the potential usefulness of phytoremediation for chlorinated solvents can be made.

016 Development of a Biological Permeable Barrier to Treat Contaminated Ground Water Using PVA-Immobilized Cells. Razavi Shirazi, F*., Oklahoma State University; Stillwater, OK; Veenstra, J., Oklahoma State University, Stillwater, OK. Today's release of chlorophenols to the ground water is greater and growing faster than ever. Due to their toxicity, tendency to bioaccumulate, and persistence in the environment, chlorophenol contamination of ground water is of concern. This research focused primarily on development of biological permeable barriers as a method of cleaning up ground water contaminated with Trichlorophenol (TCP). Permeable barriers are permeable to water, but prevent the migration of contaminants. This study investigated the performance of a novel permeable barrier system: cells immobilized in poly-vinyl alcohol-boric acid (PVA) beads. Treatment systems using immobilized microbial cells have significant advantages over conventional free cell systems. Immobilized microbial cells have been shown to be more resistant to high concentrations of toxic chemicals because the cells are entrapped in polymer a (PVA). This study evaluated the use of immobilized cells in a permeable barrier system by conducting a series of column studies under a variety of conditions, such as different flow rates and different concentrations. The permeable barrier was evaluated on the basis of removal efficiencies, ease of operation and cost. PVA-immobilized cells had TLP removal efficiencies between 100-91% for loading rates ranging from 0.3-0.6 g $L^{-1}d^{-1}$. Resilience of the PVA-immobilized cells was examined by exposing the system to high concentrations of TCP (10 times normal concentration) and low DO conditions (. 2 mg/L DO) and then tracking their recovery after normal operating conditions were restored. The permeable barriers recovered within 11 to 21 days. The cost of PVA-immobilized cells permeable barrier is very low compared to other technologies for *in situ* removal of contaminants from ground water. Overall, the PVA-immobilized cells' performance were considered to be good, making this specific matrix permeable barrier a potentially useful technology.

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daughter products resulting from reductive dechlorination of chlorinated organic compounds. The results of the geochemistry data were then used to calculate degradative rate constants. These results were incorporated into the site solute fate and transport model to further assess the effectiveness of natural attenuation of the volatile organic contaminants. These results have demonstrated that chlorinated ethenes, in this system, are subject to sequential reductive and oxidation processes that favor natural attenuation. The sequential redox zone-specific results of reductive dechlorination of site constituents of concern will be presented.

012 Identification of Bacteria in Contaminated Ground Water Using the RNA-Hybridization Technique. Farmer, J.J., Byl, T.D., Tennessee State University and U.S. Geological Survey, Nashville, TN, USA; Williams, S.D., U.S. Geological Survey, Nashville, TN; and Bailey, F.C., Middle Tennessee State University and U.S. Geological Survey. The RNA-hybridization technique is a new method to identify microorganisms. The RNA hybridization technique can identify microorganisms by taking advantage of unique genetic sequences on ribosomal RNA. This technique was used to identify bacteria in karst ground water at a site contaminated with chlorinated solvents. Well samples of anaerobic ground water had a higher percentage of sulfate-reducing bacteria and methanogenic bacteria. These bacteria are known to cause anaerobic reductive dechlorination of chlorinated solvents. Well samples of ground water that fluctuated from anaerobic to aerobic contained sulfate-reducing bacteria, ammonia-oxidizing bacteria, and methane-oxidizing bacteria. The latter two are known to biodegrade chlorinated solvents through cometabolic processes. This bacterial information was used in conjunction with geochemical and hydrologic information to identify the most prominent pathways of biodegradation at the contaminated site.

013 Evaluating BOD, COD, and TOC as a Surrogate Measure of Food Replenishment for Bacteria in a TCE-Contaminated Karst Aquifer. Akula, S., Tennessee State University, Nashville, TN; Williams, S.D., and Byl, T.D., U.S. Geological Survey, Nashville, TN. Chemical and biological data collected at a TCE-contaminated karst site in Middle Tennessee indicate that biodegradation is taking place naturally at the site. If bioremediation is to be a corrective action at the site, food replenishment and sources for microorganisms should be evaluated. Biochemical oxygen demand (BOD), chemical oxygen demand (COD) and total organic carbon (TOC) were measured in samples from several wells at the site as possible indicators of food replenishment. Water-level data and additional water-quality data such as dissolved oxygen (DO), pH, specific conductance and temperature were monitored in-situ over a 2 month period to determine how hydrologic and water-quality changes may be related to food replenishment. Continuous field data show that DO in 100 foot-deep karst conduits was replenished within a few hours after rain events. Synoptic sampling has demonstrated that water in some wells was consistently low in DO (<0.1 mg/L) and high in COD regardless of weather conditions. Though BOD and TOC concentration in samples from these same wells appeared to vary in response to changing weather conditions, TOC did not appear to be well correlated with weather patterns. The correspondence of rain events with changes in BOD shows that BOD was generally highest under dry conditions indicating a possible lag time between food replenishment and BOD response. Lab experiments conducted on field samples and spiked with ammonia and acetate verified these findings. The addition of food in lab studies also stimulated the rate of biodegradation by 350 percent over that observed in sterile controls. Lower concentrations of food stimulated the biodegradation to a lesser extent.

014 Microcosm Study to Determine the Feasibility of Bioremediation at a KarstSite. Franklin, P.R.*, Tennessee State University, Nashville, TN;Williams, S.D., Farmer, J.J., and Byl, T.D., U.S. Geological Survey,Nashville, TN. The potential for bioremediation of trichloroethylene (TCE) is being examined at a karst site in Middle Tennessee where "pump and treat" wells are used to slow the movement of ground water away from the site and to remove some of the TCE. Ground-water samples were collected from nearby wells and analyzed for geochemical indicators of bioremediation (electron donors and acceptors). Trends in the concentrations of TCE and intermediate degradation products also were monitored. Bacteria associated with the degradation of TCE were identified in water samples using the RNA oligonucleotide hybridization technique. Water-quality data indicate that both aerobic and anaerobic conditions occur in the karst aquifer. Sulfate-reducing and methanogenic bacteria that are associated with the reductive dechlorination of TCE (an anaerobic process) were present in samples of anaerobic well water. Methanotrophs and ammonia-oxidizing bacteria, associated with co-metabolic degradation of TCE (an aerobic) process), were present in samples of aerobic well water. Aerobic and anaerobic bacteria were identified in well water that fluctuated in dissolved-oxygen concentrations. Microcosm studies of ground water collected from the karst aquifer indicated that both biodegradation pathways are active. The biodegradation in some well water was rapid, with a TCE half-life of 35 days. Additional water-quality and hydrologic monitoring of the karst aquifer are being conducted to determine temporal and spatial changes in water quality and to evaluate how these changes may affect the biodegradation processes at the site.

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021 Biological Assessment of Soil, or SETAC Retraces Evolution from Water to Land In the Evaluation of Ecological Toxicity. Linder, G.,* HeronWorks, Salem, OR and USGS, ECRC, Columbia, MO; Stephenson, G., Ecological Services for Planning, Guelph, ON, Canada; Fairchild, J., USGS, ECRC, Columbia, MO. SETAC has a long history of encouraging the development of biological assessment tools applicable to ecological hazard and risk evaluation. While numerous compilations of aquatic and sediment test methods are readily available for evaluating toxicity as an ecological effect, fewer collections of test methods are readily accessible for evaluating soil contamination. Here, through a series of platform presentations, we will present numerous biological assessment tools applicable to soil contamination evaluations, with a central focus on methods that are currently available for evaluating soil toxicity. Soil toxicity is one potential indicator of adverse effects associated with soils impacted by chemicals released to the environment, and biological tests completed on soils potentially yield endpoints relevant to the evaluation of ecological effects within an ecological risk setting. Biological test methods -- animal, plant, or microbial -- that assess "upland" soils will be emphasized in the session, but methods applicable to hydric soils characteristic of wetlands will also be considered. In general, the soil evaluation methods detailed in this session will be tests on bulk soils or tests that evaluate toxicity associated with soil interstitial water. Within an ecological risk context, the biological assessment tools will focus on acute toxicity tests as well as short-term tests measuring biological endpoints characteristic of chronic effects associated with soil contamination. As such, estimates of soil toxicity may yield information regarding acute biological responses as well as suggest longer-term biological effects (e.g., genotoxicity or teratogenicity) potentially associated with subacute and chronic exposures to complex chemical mixtures characteristic of most soil exposures.

022 Comparative Toxicity of the Earthworm *Eisenia foetida* Relative to Soil Type. Simini,M.*, GEO-CENTERS, Inc. and Ronald T. Checkai, U.S. Army ERDEC, APG, MD USA. Chemical warfare agent simulant compounds (CWASC) used in testing and training exercises can contaminate soils at US Army installations. Formerly, little was known about the effect of soil type on toxicological response of ecologically important soil fauna. Evaluation of this differential response is important to estimate ecological risk. Acute (14-day survival) and chronic (21-day cocoon production) earthworm (*Eisenia foetida*) tests were employed to evaluate the toxicity of the CWASC: Bis (2-ethylhexyl) phosphonate (BIS), diisopropyl methylphosphonate (DIMP), and Malathion in standard artificial soil (AS) (U.S.EPA), O'Neil-Hall Sandy Loam (OHSL) and Sassafras Sandy Loam (SSL). Results indicated greater sensitivity to all CWASC in chronic tests compared to acute tests. Toxicity varied according to soil type. BIS was significantly more toxic in both field soils compared to the artificial soil in both acute and chronic tests. Malathion was more toxic in SSL than in AS, but less toxic in OHSL in the acute tests. Chronic test results for Malathion indicated greater toxicity in AS and SSL compared to OHSL. Toxicity was not significantly different for DIMP among all soil types for both acute and chronic tests. Soil organic matter (OM) may play a role in determining degree of toxicity. Organic matter content in AS, OHSL, and SSL was 10%, 4.3%, and 2.3%, respectively.

023 Comparison of the Bioavailability of Pentachlorophenol in Soils Using Earthworms (*Eisenia fetida*) and Passive Sampling devices. Lanno, R.P. and J. Wells, Oklahoma State University, Stillwater, OK. The soil environment contains myriad modifying factors that affect the bioavailability of toxicants to soil organisms. Critical body residues (CBRs), or chemical residues in organisms related to biological responses, provide one method of directly assessing bioavailability of chemicals from soils. However, CBR evaluations are restricted to soils where test organism health is not compromised by normal soil characteristics. An alternative to live organisms for assessing the bioavailability of soil contaminants is passive sampling devices (PSDs) such as semi-permeable membrane devices (SPMDs) and solid-phase microextraction fibres (SPMEs). This study compares CBRs for pentachlorophenol (PCP) in *E. fetida* to residues as determined by PSDs. Earthworms were exposed in artificial soil spiked with PCP in a geometric concentration series to estimate incipient lethal levels (ILLs) and toxicity half-lives. SPMDs were exposed at the estimated ILL (65 mg/kg) and sampled in a geometric time series up to 64 days. SPME estimates of PCP availability will be made at the same times as SPMD samples were taken. A comparison will be made of PCP residues in both worms and SPMDs at steady state and the implications of bioavailability as measured by PSDs for ecological risk assessment will be discussed.

024 Bioaccumulation of Metals and Selected Organic Chemicals in Earthworms. Linder, G.,* HeronWorks, Salem, OR and USGS, ECRC, Columbia, MO; Callahan, C., US EPA, Region 9, San Francisco, CA; Fairchild, J., USGS, ECRC, Columbia, MO. As part of the ecological risk assessment process for contaminated soils, the evaluation of bioaccumulative chemicals is frequently pursued through a food-chain analysis focused on exposures to terrestrial vertebrates followed by a comparison of daily doses to benchmark toxicity reference values to estimate risk. Quite often, however, a quantitative description of *soil-to-soil biota* transfer factors is lacking. Out of necessity then, simple ratio estimators are taken from the existing literature to estimate tissue residues in prey items of terrestrial vertebrates. For example, earthworms most often are considered *vectors* in the transfer of soil contaminants to receptors in higher trophic levels; most often, birds and mammals. Here, designed studies focused on food-chains such as "soil → soil macroinvertebrate →terrestrial vertebrate" illustrate an approach for the derivation and validation of trophic transfer factors for metals such as cadmium, chromium, copper, lead, and zinc, which are frequent constituents in contaminated soils. Similarly, an analysis of the bioaccumulation potential of selected organic chemicals will be summarized, including an analysis of the confounding effects associated with earthworm foraging during exposure. Overall, the results of these studies suggest that food-chain analysis need not rely exclusively on *encountered data* to estimate the transfer of chemicals from abiotic to biotic compartments, especially given the critical role of transfer factors in the analysis and the potential for their being influenced by numerous interacting abiotic and biotic factors such as the physicochemical properties of soil and the biological responses of earthworms during exposure.

025 Biological Assessment of Soils by Using a Chronic Plant Test. Römbke, J.*, Kalsch, W., ECT Oekotoxikologie GmbH, Flörsheim, Germany. Since 1994, the German government is funding a joint research project entitled *Processes for the Bioremediation of Soil.* One part of this initiative is the validation and (partial) development of more than ten ecotoxicological test methods as a complement to chemical analysis. These methods are considered to be used in the future for the assessment of the retention function (ability of soils to adsorb pollutants in such a way that they cannot be mobilised via the water pathway) and habitat function (ability of soils to serve as a substrate for plant growth and as a basis for biocoenosis) of bioremediated soils. After an overview on the methods currently under study in this project, a chronic plant bioassay conducted in our laboratory will be used as an example. This new test is based on experiences with various plant acute tests (especially the OECD *Growth Test* (1984)) and the

Plant-Life-Cycle Bioassay developed in Canada in 1993. Characteristic properties of the test system, which can be performed either with *Brassica rapa* or *Avena sativa*, are the duration of 35 to 50 days, the use of the OECD artificial soil and a German standard field soil as controls, an *automatic* wick irrigation system and, besides measuring biomass and shoot length, the number of pods, seeds and flowers are used as measurement endpoints. During the development of the test, TNT was used as a reference substance. The results of these test runs, e.g. an NOEC of 56 mg/kg in a test with *B. rapa*, are compared to tests in which contaminated and uncontaminated soil samples from an old ammunition site (TNT concentration: up to 1600 mg/kg) were tested before and after bioremediation.

026 Relevance of Laboratory Toxicity Tests for Risk Assessment of Soil Contamination. Smit, C.E.*, Posthuma, L., National Institute of Public Health and the Environment, Bilthoven, The Netherlands; Van Gestel, C.A.M., Vrije Universiteit, Amsterdam, The Netherlands. Results of single-species laboratory toxicity experiments play an important role in the derivation of soil quality criteria in the Netherlands, since they are the input values for the statistical extrapolation methods used to estimate ecotoxicological risk limits. The project "Validation of Toxicity Data and Risk Limits for Soils" aimed to evaluate the field relevance of both toxicity data and risk limits, and to identify which factors introduce the largest uncertainties in laboratory-to-field extrapolation of toxicity data. Experimental research focused on metal contamination, in particular zinc. Laboratory tests and semi-field experiments on widely used test species (*Trifolium pratense, Folsomia candida, Enchytraeus crypticus, Eisenia andrei)* and micro-organisms were performed to determine the influence of soil characteristics and exposure conditions on zinc toxicity. It appeared that variable exposure conditions modulate toxicity only in a minor way, whereas differences in bioavailability between soils are of prime importance in laboratory-to-field extrapolation. It was shown that soil extraction with water or a $CaCl_2$ solution can be used as an operational measure of bioavailability. To study the ecological relevance of extrapolated risk limits, observations on community endpoints of enchytraeids, nematodes and micro-organisms were made in an artificially contaminated field plot and at a heavy metal polluted field site. Results indicate that the current Dutch ecotoxicological hazard concentrations discriminate between the absence and presence of contaminant effects in the field. These risk limits are therefore valuable as a first-tier evaluation system in soil protection policy. For site-specific risk assessment, the soil chemical and biological characteristics of the local ecosystem should be included in the evaluation.

027 Methods to Assess the Impact of Human Activities on Soil Microbial Ecosystems. Siciliano, S.D.* University of Saskatchewan, Saskatoon, SK, Canada. There are a variety of human activities such as point source pollution from smelters or the introduction of engineered organisms, that can adversely affect soil microbial communities. Traditional methods in microbial ecology designed to detect changes in soil communities are time consuming and imprecise. Newer methods such as fatty acid analysis, carbon substrate utilisation or genomic analysis, show considerable promise to improve the speed and precision of soil microbial toxicity tests. Fatty acid analysis of soil can detect changes in soil microbial communities caused by heavy metal or chlorinated benzoic acid pollution. However, background variation due to a variety of soil properties such as sampling depth, landscape position or soil type, can confound the results obtained with toxicity tests. Hence, careful selection of controls is required to insure that results are interpreted correctly. Assessing the carbon substrate utilisation patterns of microbial communities extracted from soil can also assess the impact of pollution but relating these profiles to larger ecosystem processes is difficult and requires further research. Furthermore, comparisons between studies is hampered due to poor control of inoculum density which is known to influence utilisation patterns. Extraction of DNA from soil and the analysis of genomic sequences is a proven technique but suffers from technical difficulties. Humic acid polymers co-extracted with DNA, interfere with many nucleic acid techniques and makes processes such as DNA amplification difficult. This technique, however, gives readily interpretable results that can be related to larger ecosystem processes. These new tools promise accurate and cost effective measurements of the impact of human activities on the soil microbial community.

028 Assessing the Ecological Risk of Soil Contaminants in Remedial Actions - Perspectives from the Trenches. Stahl, R.G.*, DuPont, Wilmington, DE; Blakley, N.R., Washington Dept. of Ecology, Olympia, WA. Assessing the ecological risk of soil contaminants presents an important challenge to practioners of ecological risk assessment (ERA) as well as to individuals and groups tasked with making a final risk management decision. The challenge is common to those in the regulatory and regulated communities alike. Two problems occur routinely: 1) even though soil contaminants have been evaluated in detail for assessing risks to humans, the same methods and approaches are not applicable, fully, in ERA; and, 2) unlike the approach to developing water quality criteria, where data sets are relatively robust, similarly large and diverse data sets are not routinely found for terrestrial receptors. Common challenges in undertaking the ERA include: 1) what studies to select to support the development of a toxicity reference value (TRV); 2) finding suitable, scientifically-sound data to support inputs to food web models including soil to tissue uptake factors, contribution of soil to dietary intake, relevant biomarkers of exposure and effects, and , a host of other issues. Using soil toxicological test results and other relevant biological information, allows risk assessors and risk managers to reach a scientifically supportable remedial decision.

029 Biological Assessment of Hydrocarbons in Soils. Menzie*, C.A., Menzie-Cura & Associates, Chelmsford, MA. This paper outlines a tiered approach for evaluating hydrocarbon-contaminated soils. The approach reflects experience with petroleum as well as coal-tar contaminated sites. The approach begins with a screening-level method that helps identify if ecological risk assessment is warranted. This initial step is also used to begin the process of identifying what is important to protect at a site and to identify the important pathways. Information developed in this initial screening-level analysis is then used to identify contaminated media/pathways that are relevant to the site. The next step of the procedure involves the use of screening criteria that may include *benchmarks* as well as site observations. Biological assessment tools including field observations and measure of toxicity to plants and animals are included in subsequent tiers. The method describes how spatial and temporal scales can be considered for both the exposure field as well as for the activity patterns of the ecological receptors. Finally, information is provided on how to include measures of chemical availability into an overall weight-of-evidence approach.

030 It's Soil, Not Mud! What We Know, Assume, and Rationalize on Our Path to Assessing Terrestrial Systems. Kapustka, L. A., ecological planning and toxicology, inc., Corvallis, OR. Contemporary approaches to ecological risk assessment are dominated by estimates of toxicity. Policies adopted by regulatory bodies were based largely on experiences with aquatic systems. In addition, most environmental quality criteria endorse precautionary principles to finesse issues of uncertainty. Many state, provincial, and international regulatory bodies are marching rapidly toward the adoption of soil criteria for organic and inorganic substances. Meanwhile, humbling questions are being asked, which call into question whether the scientific support exists to set discrete soil criteria. The literature is awash with reports from hydroponic studies and other experimental designs that hardly reflect the dynamic nature of soils. This paper will highlight the multi-factor, non-linear relationships that dominate phytotoxicity and ecological assessment of soils, including: relationships between environmental concentration and plant tissue concentration; comparisons among mycorrhizal and non-mycorrhizal plants and gnotobiotic plants versus plants with rhizosphere flora; and finally translation of physiological response to toxicological effect to ecological consequence. It won't be pretty; it's soil.

031 What Is the Optimum Number of Replicates and Number of Observations per Replicate for the Sea Urchin Embryological Development Assay? Mastroti, R.R.*, University of Sao Paulo, Sao Paulo, SP, Brazil; Rosso, S., University of Sao Paulo, Sao Paulo, SP, Brazil. Toxicity tests for monitoring purposes should be sensitive and easy to conduct. Monitoring programs often generate a high number of samples due to the numerous sampling sites and sampling frequency. The goal of this study was to evaluate how the number of replicates and the number of organisms observed per replicate influenced the results. With this knowledge it becomes possible to minimize the observation efforts without impairment to the reproducibility and reliability of the test. A short term chronic toxicity test was conducted with sea urchin embryos (*Lytechinus variegatus*). Ten replicates were prepared for each of the six selected concentrations of SDS (reference toxicant). The percentage of abnormalities for each replicate were calculated on the basis of 20, 40, 60, 80, 100, 120, 140, 160, 180 and 200 organisms observed per replicate. The variances and averages associated with the number of replicates and with the number of observed organism per replicate were calculated. Based on this analysis, some advantageous combinations were suggested to be applied in routine toxicity tests.

032 Estimation of Hazardous Concentrations from Bimodal Species Sensitivity Distributions Using Bayesian Statistics. Jaworska, J.S.*, Procter & Gamble, Strombeek-Bever, Belgium; Aldenberg, T., RIVM, Bilthoven, The Netherlands. The sensitivity of biological species for toxicants has been modeled through fitting unimodal statistical distributions to NOEC, or LC50, data sets. Quality objectives can be calculated by estimating percentiles, so-called Hazardous Concentrations, at which a fraction of 5% of the species is affected at the most. For estimating the Fraction Affected at some given or predicted environmental concentration, the method is used inversely by calculating values of the cumulative sensitivity distribution. Since toxicity data sets tend to be small, the estimation of the uncertainty, either through Classical confidence statistics or Bayesian statistics, is an essential part of the assessment process. For pesticides, unimodal distributions may not be appropriate, since many pesticides are developed for certain target species, so that the toxicity data may be distributed in a bimodal or multi-modal way. Using a single mode distribution including data for insensitive species may lead to an overestimation of the risk involved. One way to model bimodality is to employ mixtures of two unimodal distributions. The simplest model involves five parameters: two means, two standard deviations, and a weighting factor. The Bayesian approach is flexible enough to handle the fitting of these bimodal distributions to data sets, as well as to assess the uncertainty of the distribution employed, of its cumulative function and of the percentiles to determine Hazardous Concentrations. Since the mixtures involve a more sensitive group (lowest mean) and a less sensitive group (highest mean), one has at least two options to set Hazardous Concentrations, either as a percentile of the total mixture, or as a percentile of the more sensitive group. This latter option is preferred. For fenitrothion L(E)C50 data (n=42), clearly showing bimodal behavior, it turns out that the several possible options for estimating the median HC5 do not differ much: 0.30 ug/l from the unimodal normal distribution, 0.26 ug/l from the unimodal model for the most sensitive data only, 0.37 ug/l from the bimodal normal mixture, and 0.16 ug/l from the more sensitive group of the bimodal mixture. This is surprising given the fact that the data span a range of 0.021 up to 7000 ug/l. The cause of this apparent insensitivity to the model employed is discussed.

033 Statistical Analysis of Ecotoxicology Experiments: Practice, Relevance and Power. Green, J. W., DuPont, Newark, DE. Several methods of analyzing continuous data in a concentration-response experiment will be compared for power and appropriateness, including Dunnett's classic multiple comparison procedure and step-down applications of the Jonckheere-Terpstra trend test and a new contrast based approach. Two underlying principles are recommended: (1) The statistical model should reflect the experimental design and both biological expectation and uncertainty. (2) Demonstration of adequate statistical power should be a requirement for every analysis. In general, a step-down approach is recommended for monotonic data with an appropriate check of the presumptive model. In this context, contrasts using normalized ranks and Bartholomew's test for trend or departure from trend are compared. Simulation results and laboratory test data are used to demonstrate the power and application of two possible statistical tests consistent with these recommendations and the tests are compared to Dunnett's test. Although the Jonckheere-Terpstra test is well known, the step-down application of this test is relatively new. The theoretical basis of this approach and simulation results are given. An alternative test is based on Helmert contrasts and was developed by A. C. Tamhane, C. W. Dunnett, J. D. Wetherington and this author. This latter method is especially appropriate when agreement exists on the magnitude of biological effect it is desirable to detect. As a rule, step-down tests based on monotonic response models are more powerful than multiple comparison methods which assume no structure. The use of one- or two-sided tests depends on biological appropriateness. The variety of conditions of normality, variance homogeneity, multiple variance component designs, lack of monotone concentration-response and presence of massive ties in the response found in ecotoxicology data mean none of these methods is universally appropriate, but uniform, logical flow charts exist.

034 No Observed Effect Concentrations (Noecs) and Effect Concentrations (Ecs) for Identifying a Safe Concentration. Green, J.W., Wetherington, J.D.*,Ferry, N.M., Dupont Agricultural Products, Newark, DE. The primary objective in aquatic toxicology laboratory studies is to assess the biological activity of a chemical compound to nontarget aquatic animals (e.g., fathead minnows). An experiment is generally conducted in which five or more concentrations of a compound are administered to separate groups of experimental units. The goal of these experiments is to estimate a safe concentration that will not produce some undesirable level of biological effect (e.g., reduction in egg production in female fathead minnows). There are two general approaches to estimating a safe concentration. One approach uses hypothesis testing to find the highest concentration that is safe among the tested concentrations. This is the highest concentration at which the mean response is not statistically significantly different from the mean response of the zero concentration control group (i.e., the No Observed Effect Concentration or NOEC). A second approach for estimating a safe concentration uses regression methods to describe the relationship between concentration and response with a mathematical function (e.g., a line). Using this hypothesized relationship, the concentration corresponding to a specified level of effect is predicted (e.g., the Effect Concentration producing a 5% reduction in egg production or EC_5). An increasing number of statisticians favor EC estimation over the calculation of NOECs. First, we review the criticisms leveled against determining NOECs and present data to indicate that these criticisms are not necessarily fatal. Second, we compare conclusions drawn from NOECs calculated from actual aquatic toxicology experiments to conclusions drawn from predicted ECs and demonstrate the dependence of the predicted EC on the choice of mathematical function. Finally, we introduce an alternative approach for identifying a safe concentration based upon step-down multiple comparisons procedures (e.g., Biometrics 52: 21-37).

035 Hockey Stick and Generalized Linear Models (Glims): How Useful Are They for Estimating Thresholds? Moore, D.R.J.*, The Cadmus Group, Ottawa, ON; Caux, P.-Y., Environment Canada, Hull, PQ. One objective of analyzing toxicity data is to derive the level below which there are no adverse effects (*i.e.*, the threshold). Proponents of hypothesis testing often argue that the regression approach using standard models (probit, logistic) precludes threshold estimation, thus justifying using ANOVA to estimate NOELs. This is not a valid defence of ANOVA, but is a valid criticism of the regression approach. In this paper, we explore and critique two regression approaches capable of estimating thresholds: generalized linear models (GLiMs) and hockey stick models. Bailer and Oris (ET&C 16: 1554-1559) demonstrated that GLiMs provide a general framework for data commonly encountered in aquatic toxicology including survival, fecundity, and biomass. GLiMs have two ingredients: the distribution of the response variable (*e.g.*, binomial distribution for survival), and a link function to linearize the concentration-response relationship (*e.g.*, probit link for survival). By adding a quadratic term to the linear regression equation, GLiMs can be used to estimate threshold concentrations. Hockey stick models are a special case of the segmented class of regression models. They consist of two segments, both linear, constrained to intersect at the threshold. One segment has a zero slope to represent constant background effect. The threshold and the slope of the other segment are the estimated parameters. We found that both approaches are capable of estimating thresholds, although several treatments with no effects and several with low toxic effects are generally required to estimate thresholds with reasonable precision. Hockey stick models have the added advantage of being able to estimate confidence intervals for the threshold. GLiMs, however, can handle hormetic responses and have more theoretical justification than do hockey stick models.

036 Identifying and Quantifying Hormetic Hazards. Bailer, A.J.*, Oris, J.T. Center for Environmental Toxicology and Statistics, Dept. of Math. & Stat.(AJB), Dept. of Zoology (JTO), Miami University, Oxford, Ohio USA. Subtoxic stimulation of organism response is not uncommon in aquatic toxicology experiments. This subtoxic stimulation, so-called hormesis, is observed in both growth and reproduction experiments in different trophic levels of an ecosystem. The quantification of hormetic hazards is described. In particular, inhibition of response relative to control response or relative to maximal response will be discussed. The use of statistical models that accomodate subtoxic stimulation is advocated from the perspective of potency estimation. Finally, we describe issues in the design of toxicology experiments in which hormesis is expected.

037 Technical Basis and Application of Toxicity Equivalents in Probabilistic Risk Assessments of Pesticide Mixtures. Chappel, M.J.*, Solomon, K.R., Sibley, P.K., Centre for Toxicology, University of Guelph, Canada. Understanding the toxicology and risk assessment of multiple stressors has historically been a challenge in environmental toxicology. However, in reality, most biological systems are exposed to multiple stressors at the same time. For compounds which exhibit the same mechanism of toxic action, effects will most likely be additive. Where additivity exists, Toxic Equivalency (TE) approaches may be used as basis for the risk assessment of these compounds. An initial concentration-response study followed by an ANOVA study were used to determine whether the toxic equivalency (TE) approach could be used for probabilistic risk assessments for mixtures of organophosphorus insecticides. In our study, the effects of mixtures of three organophosphorus insecticides (azinphos-methyl, chlorpyrifos, diazinon) on populations of fathead minnows (*Pimephales promelas*) were evaluated in both field mesocosm ($12m^3$) and controlled laboratory studies. In the ANOVA study, the three OP's were normalized to the most toxic organophosphorus insecticide, in our case, azinphos-methyl. For the field mesocosm studies, the exposure values were based on an ecosystem-level 90^{th} centile response derived using distributional analysis of existing measured environmental exposure data sets. In both studies, mortality and brain acetylcholinesterase (AChE) activity were monitored for up to 7-d. In the concentration-response study, significant reductions in survival of fish were observed at 150 x 90^{th} centile, equaling approximately 51 mg/L azinphos-methyl equivalents. When tested in the ANOVA study, both mortality and brain AChE activity were not significantly different between each treatment, indicating that concentration addition was the only interaction observed. This study validates the use of TEs for the probabilistic risk assessment of mixtures of organophosphorus insecticides and confirms the additivity theory for the effects of mixtures of organophosphorus insecticides. Preliminary laboratory studies also confirm the additivity hypothesis and will be presented.

038 Importance of Understanding Sensitivity in Model Ecosystems: Relationships to Field Communities and Risk Assessment Implications. Belanger, S. E. and Dyer, S. D. The Procter & Gamble Company, Cincinnati, Ohio USA. Well designed model ecosystems are used as surrogates for the real world in evaluating the fate and effects of chemicals. Benthologists have long recognized that some fauna are more susceptible to effluents than others. These include the Ephemeroptera, Plecoptera, and Trichoptera (EPT) group and selected members of almost all other invertebrate orders. Low application factors are assigned to results from model ecosystems for risk assessment purposes. This implies that the experimental design was suitable to find responses when present and that the biology was of a sufficient type (containing sensitive species and sufficient abundances) to detect perturbations. In order to quantitatively assess this issue we first evaluated the community structure of numerous model stream ecosystems relative to field communities. Secondly, we compared results of a single model stream system (the P&G Experimental Stream Facility) which can use effluent and/or surfactants as perturbants with a long term field survey of sites throughout Ohio conducted by Ohio EPA. In the first analysis, we demonstrate that model ecosystems have the potential to simulate community complexity equivalent to that of impacted and unimpacted field communities. Statistical sensitivity (minimum detectable difference, coefficients of variation) for selected designs and test systems was also determined. In the second, the ESF is placed into the context of community sensitivity of Ohio rivers using population sensitivity ranking indices based on work of Clements (1992), Hilsenhoff (1987) and Lenat (1993). The ESF is in the upper 20 percentile of stream communities with respect to sensitivity. These results quantitatively support the use of low or no assessment factors for extrapolating results of sensitive model stream ecosystems for use in risk assessment.

039 a Strategy for the Use of Multivariate and Multimetric Methods of Analysis of Benthic Macroinvertebrate Communities to Assess Different Types of Stress. Boyle, T.P.*, Biological Resources Division, USGS, Fort Collins, CO; Easter, M.J. Colorado State University, Fort Collins, CO. The use of community level biological assessment has become divided into two approaches. The first has come to be know as the multimetric method in that it is comprised of a number of metrics on individual community attributes which are then presented as an array and/or summed to give a single measure of the status of the biological community. The second is the use of various multivariate statistical techniques, which assess the structural variability of the benthic macroinvertebrates and the relationship of that variability with natural and anthropogenic environmental variables. The multimetric methodology has the advantage of ease of computation and comprehension; the multivariate methodology, while computationally more complex, is more statistically rigorous and better integrates physical, chemical, and biological data. Using canonical correspondance analysis, we have developed a rational for the tandem use of these two methodologies and offer example studies of changes within benthic macroinvertebrate communities at different sites with different degrees and types of stress, including landuse (sedimentation) in ther Upper Pennsula of Michigan, abandoned mine lands (heavy metals) in Montana, and domestic treatment plant effluent (ammonia) in southern Arizona. Based on the ordinal separation of the different sample sites within a watershed, and the strength of the relationships of the analysis with the physical and/or chemical variables, individual metrics were selected or developed for future assessment and monitoring.

040 A Framework for Assessing Contributions of Multiple Stressors to *in-situ* Biological Responses. White-Hull, C.E.*[1], S. Dyer[1], X. Wang[2], T. Johnson[1], and G. Carr[1]. [1]The Procter & Gamble Co., Cincinnati, Ohio and [2]Univ. of Cincinnati, Cincinnati, Ohio. Conventional assessment methods estimate risk to receiving water biota via univariate analyses of laboratory-derived biological information and chemical exposure data, and assume that biological response can be predicted based upon an adequate understanding of single chemical exposure. Although this approach is suitable for single species toxicity tests and extrapolation to model ecosystems, it may be unsuitable for field situations where other physical and chemical factors (e.g temperature, suspended solids, in-stream and riparian habitats, pH, nutrients, and chemicals from agricultural, consumer and industrial uses) may play more important roles in regulating biotic integrity. Therefore, in order to ground-truth lab-based risk assessments, an understanding of the relative contributions of these other factors dictating biotic integrity should be considered. This presentation will describe a framework developed for assessing these contributions based upon a pilot study conducted for the Little Miami River watershed, an Exceptional Warm-Water Habitat located in southwest Ohio. For the approach, water chemistry, habitat and biological information are extracted from Ohio and U.S. EPA databases, and integrated via Geographical Information System (spatial analysis) technology using a unique stream segmenting system. The importance of water chemistry and habitat are then determined using multivariate statistics. Based on the success of the pilot study, the approach was applied to additional Ohio watersheds having more diverse habitat and water chemistry characteristics. The presentation will address key methodology issues: data stratification (e.g. watershed, sub-watershed, drainage area), scale of data aggregation (e.g. river segmentation, nearest neighbor approach), and statistical considerations. Results using the framework for these additional watersheds will also be presented.

041 The Immunophysiology of The Gulf Killifish, *Fundulus grandis*: a Model for Assessing Estuarine Environmental Health. Rice, C.D.*, Banes, M.M., Andol, A.Z. CEHS, College of Veterinary Medicine, Mississippi State University, P.O. Box 9825, Mississippi State, MS 39762, USA. The gulf killifish, Fundulus grandis, is common in salt marshes along the northern Gulf of Mexico, USA. The home range of this fish is limited, consequently it is chronically exposed to local environmental conditions, including pollution and pathogenic stress. As with the mummichog, F. heteroclitus, a very closely related species from the east coast of USA, F. grandis adapts well to laboratory rearing practices and is routinely used in reproductive, developmental, carcinogenesis, and chemical adaptation studies. However, little is known of it's immununophysiology beyond innate immune responses measured *in vitro*. We developed monoclonal antibodies (mAb) against circulating and lymphocyte-bound immunoglobulins (Ig) of F. grandis that, in turn, allows us to measure antibody responses of this fish. We also developed a mAb specific for eosinophilic granular cells (EGC), the primary lymphoid-associated phagocyte in the genera Fundulus. Using these mAbs and leukocyte function assays, we find that (1) F. grandis mount vigorous antibody and cellular immune responses to Vibrio anquillarum, a prevalent marine pathogen in many polluted estuaries, (2) there are circadian and seasonal changes in total Ig and antigen-specific Ig, and (3) that 10, 1, 1, and 10 ppm of aroclor 1254, TBT, 3-MC, and nonyl-phenol, respectively (in mixture as a 120 day dietary exposure), alters circadian and seasonal changes in total and antigen-specific antibody immune responses. In addition to affecting circadian and seasonal changes in immune responses, the treated diet induces immunoreactive hepatic microsomal CYP1A protein, however this biomarker also exhibits circadian and seasonal changes in both treated and control fish. (Funded by MS-AL SeaGrant NA16RG0129-R/ER-37PD)

042 Effects of Exposure to Pentachlorophenol on Host Resistance, Nonspecific and Specific Immunity in the Bluegill (*Lepomis macrochirus*). Beaman, J. R.*, GEO-CENTERS INC., Ft. Detrick, MD; Hoffmann, F., Finch, R. A., & Gardner, H., USACEHR, Ft. Detrick, MD. The effect of pentachlorophenol (PCP), a polychlorinated fungicide/pesticide, on immune function was studied in a common freshwater teleost, the bluegill (*Lepomis macrochirus*). PCP is rapidly absorbed through inhalation, ingestion, or through dermal exposure. PCP uncouples oxidative phosphorylation, acts as an enzyme inhibitor, and stresses metabolic activity. Two 21-day flowthrough exposure studies were conducted. The first exposure study utilized juvenile (~ 6-8 months old) bluegill exposed to PCP at 0.027 and 0.110 mg/L. Chemically exposed and clean-water control groups were challenged by intraperitoneal inoculation with 1.2×10^7 CFU (~LD_{50} dose) of *Yersinia ruckeri* on days 10 and 21 of exposure in order to assess host resistance against a bacterial pathogen. Mortality incidence was monitored for 96 h in both the exposed and control groups, and toxicant effects on host resistance capacity was determined. A second exposure study utilized adult (~ 16-18 months old) bluegill exposed to PCP at 0.035 and 0.140 mg/L. At the completion of the exposure periods (day 10 and 21), a battery of *in vitro* tests measuring immune parameters (hematology, organosomatic indices, immune organ cellularity) and functions (reactive oxygen intermediate [ROI] production, mitogen-induced lymphocyte proliferation, antibody-forming cell (AFC) response to formalin-killed *Yersinia ruckeri*, serum agglutination, and serum immunoglobulin quantitation) was conducted on individual fish. Exposure of bluegill to increasing concentrations of PCP on days 10 and 21 indicated a decrease in the host resistance capacity ($F = 25.29$; $p = 0.0005$; and $F = 12.81$; $p = 0.0050$ respectively). *in vitro* immune function was also significantly affected by PCP exposure. The response of AFC's to formalin-killed *Yersinia ruckeri*, as well as ROI production, was decreased following exposure to PCP. Results of this study demonstrate the applicability of these immune assays to assess immunomodulation in a teleost fish model following exposure to a common xenobiotic pollutant.

043 Measurements of Complex Mixtures by *in vitro* Bioassay: Interactions of Dioxins, Furans and Polychlorinated Biphenyls (Pcbs). Sheri L. Lydick*, University of Maine, Orono; ME; Donald R. Hague, Maine Department of Environmental Protection, Augusta, ME; Rebecca J. Van Beneden, University of Maine, Orono, ME. The Penobscot River in central Maine receives a significant contaminant load from two bleached-Kraft pulp mills. Dioxins, coplanar polychlorinated biphenyls (PCBs), and other related polyhalogenated aromatic hydrocarbons present in the Penobscot River are persistent hydrophobic compounds whose toxic effects are mediated via the aryl hydrocarbon receptor (AhR). An *in vitro* bioassay has been utilized in our laboratory, which uses the mammalian cell line Hepa.luc1.1 containing pGud.luc1.1, an expression vector with dioxin-responsive elements (DREs) cloned upstream of the luciferase reporter gene. The ligand-activated receptor binds to the DRE and initiates luciferase expression. The bioassay allows quantitation of toxic equivalents (TEQs) and enables us to correlate exposure with histopathological effects. Significant levels of dioxins, furans, and PCBs have been detected in whole fish samples using this bioassay. Preliminary data suggest that the toxicity of samples containing complex mixtures of contaminants is a non-additive interaction. A two-fold increase in the amount of teleost whole-body extract, containing dioxins, furans and PCBs, resulted in a six-fold increase in the observed concentration. Several PCB congeners appear to exhibit antagonistic interactions with the AhR. Preliminary data revealed an 88-fold decrease in the observed versus expected TEQ of a mixture containing coplanar (toxic) and tetra-ortho (nontoxic) PCBs. Further studies of interactions of PCBs and dioxin/furans are currently being performed in our laboratory. Molecular analysis of the interactions of PCBs with dioxins coupled with histopathological evaluation of the animal samples tested can provide valuable information about the toxicity of these compounds.

044 Toxicological Interactions and Mechanisms of Chlorpyrifos, Dieldrin, and Methyl Mercury Mixtures. Steevens, J.A.* and Benson, W.H.,The University of Mississippi, University, MS. Chemical interactions of chlorpyrifos, dieldrin, and methyl mercury were characterized using dose response curve and graphical analyses as additive, independent, synergistic, or antagonistic. Experiments in which survival and growth of *Hyalella azteca* were assessed indicated that chlorpyrifos and methyl mercury acted additively, while dieldrin and methyl mercury, or dieldrin and chlorpyrifos interacted independently. No statistically significant interactions were observed in the bioaccumulation of the three compounds throughout a 96-hour uptake experiment. Mixture interactions of the three bioaccumulative chemicals were investigated utilizing biochemical measurements reflecting the mechanisms of action of the individual toxicants. A significant dose-dependent inhibition of acetylcholinesterase activity was observed in the chlorpyrifos treated (0.04, 0.2, and 0.4 ppb) groups (95, 45, and 11 percent of control) but not in the methyl mercury treatments (10, 50, and 100 ppb). However, methyl mercury antagonized the effects of chlorpyrifos mediated acetylcholinesterase inhibition at the lower concentrations (10 and 50 ppb) in a binary mixture. Methyl mercury and chlorpyrifos did not induce oxidative damage through protein oxidation, lipid peroxidation, or reduction in thiol content. Glutathione-S-transferase activity was significantly decreased ($p < 0.05$) to 41 % of control levels at the 0.4 ppb chlorpyrifos treatment. Mechanisms responsible for the observed interactions of methyl mercury and chlorpyrifos potentially exist within the cholinergic pathway or through alterations in metabolism. Dieldrin and methyl mercury interaction was independent, however, disruption of ion regulation was observed with both compounds at 45 and 75 ppb, and 50 and 100 ppb, respectively. The demonstrated inhibition of the enzyme Na/K ATPase, may explain the interaction observed at the whole organism level. Results of these experiments demonstrate the *real-world* interactions and toxicological effects of environmentally relevant pesticide and metal mixtures.

045 Effects of Malathion on Caudal Fin Regeneration in the Fathead Minnow (*Pimephales promelas*). Wallin, J.M.*, Exponent, Landover, MD; Atwood, H.L., Clemson University, Clemson, SC; La Point, T.W., Texas Tech University, Lubbock, TX; Tomasso, J.R., Clemson University, Clemson, SC; Anderson, T., Texas Tech University, Lubbock, TX. Pesticide and heavy metal exposures have been shown by other researchers to affect fin regeneration, body mass, and growth in fishes. A fundamental question for toxicological research is the relative sensitivity of these parameters. Juvenile *Pimephales promelas* (fathead minnow) were used to investigate the effect of organophosphate exposure on caudal fin regenerative ability. Malathion, a commonly used insecticide, was chosen for this research. Parameters measured were length of caudal fin regenerate, body length, and weight. Experiments to determine daily loss of malathion from bioassay chambers by aeration and efficiency of the analytical extraction procedure were also performed. Subchronic 24-day static renewal exposure to nominal 0.5 and 1.0 mg/L concentrations (5.8 and 11.6% of LC_{50}) of malathion were found to significantly inhibit caudal fin regeneration in *Pimephales* compared to acetone controls. An average body mass loss of 18.3% in the 1.0 mg/L treatment was also significant when compared to average body mass change in the clean water control treatment. Body growth, although inhibited in malathion-treated fish, was not statistically different among the treatment groups due to high variability in the measurements. Two-factor analysis of variance indicates that tail cutting does not significantly impact *Pimephales* weight or growth under these experimental conditions. Regression analyses indicate that exposure chamber aeration does not significantly decrease concentrations of malathion over 24 hours. In conclusion, inhibition of caudal fin regeneration was a more sensitive indicator of malathion toxicity than either weight loss or inhibition of body growth.

046 Comparison Among Three Invertebrate Species of the Body Residue for Lethal and Sublethal Effects. Landrum, P.F.* Great Lakes Environmental Research Laboratory, Ann Arbor, MI; S.W. Fisher, S. Chordas, H. Hwang, and P. Tiwari, Department of Entomology, Ohio State University, Columbus, OH. Several recent workshops called for development of residue-effects data to improve interpretation of bioaccumulation data. Three invertebrate species were exposed via food to DDE, and several PCB congeners. Since the effects data was based on body residue, the route of exposure was not critical. The lethality at 10-d for *Chironomus riparius* (1.4 mmol kg^{-1} average) required essentially the same body residue as *Hyalella azteca* (1.1 mmol kg^{-1} average). For *Lumbriculus variegatus*, no mortality could be induced with a 10-d exposure. However, lethality at 35-d was similar to that for the other two organisms 10-d response (1.0 mmol kg^{-1}). Sublethal endpoints required much lower body residues. For *C. riparius* and *H. azteca*, fecundity was affected in the range of 0.02 - 0.03 mmol kg^{-1} approximately 50 fold lower concentrations than lethality at 10-d. *L. variegatus* was less sensitive than either the midge or the amphipod with growth and reproduction affected at 0.3 - 0.6 mmol kg^{-1} across the range of compounds. Differences between species are, as expected, quite variable reflecting the general sensitivity to narcotics but in general the differences among compounds within a species was far less variable. By evaluating the responses based on organism residue, the variability was far less than evaluating the responses based on environmental concentrations. Using organism residues explicitly addresses both differences in bioaccumulation potential and the complication of multiple routes of exposure. This approach has the potential to not only permit better interpretation of bioaccumulation data but also to address exposures to mixtures of contaminants.

047 The Use of Caged Eels (*Anguilla* spp.) to Assess Chemical Accumulation and Effects in New Zealand's Largest River. Jones, P.D.*, Leathem, S.V., ESR (Institute of Environmental Science and Research), Lower Hutt, NEW ZEALAND; Huser B., Empsom, P., Environment Waikato, Hamilton, NEW ZEALAND. New Zealand's Resource Management Act (1992) requires the "effects based" monitoring of the impacts of discharges to the environment. While feral fish surveys are of use in this process there can be uncertainties in time and extent of exposure for wild fish. We have assessed the utility of caging freshwater eels in rivers to evaluate the accumulation and effects of environmental contaminants. Eels were caged at seven locations in the Waikato river for up to three months. After deployment, eels were analysed for a range of contaminants including dioxins, PCBs, metals and pulp and paper related compounds. To better assess the utility of this procedure we measured a number of biochemical parameters in the caged fish to assess possible impacts of caging stress. Biochemical indicators of stress were not altered after three months caging at control locations. This suggests that eels are amenable to caging and that observed effects are related to environmental contamination. Indicators of contaminant stress were higher in the lower sections of the river. Several indicators of stress increased at one location. This location also showed the highest accumulation of contaminants. Contaminant concentrations after three months of caging were similar to concentrations in feral eels from the same locations.

048 PAH-metabolites Identified in the Bile of Fish (Longnose Sucker and Rainbow Trout) Collected Downstream of a Condensate Spill. Birkholz, D.A.*, D. Johnston, Enviro-Test Laboratories, Edmonton, AB. Canada and A. Bollo-Kamara, Alberta Environment, Edmonton, AB. Following a spill of condensate into the aquatic receiving environment samples of fish bile were collected from longnose sucker (*Catostomus catostomus*) and rainbow trout (*Oncorhynchus mykiss*) upstream and downstream of the spill site. Bile samples were composited for each species and enzymatically hydrolyzed, extracted, acetylated, silylated and analyzed using gas chromatography/mass spectrometry. Acetylation prior to GC/MS analysis generated unique mass spectra which allowed us to distinguish alcohol and phenol metabolites from one another. Silylation prior to GC/MS analysis does not allow one to distinguish phenol from alcohol metabolites. The major metabolites found to be present in the bile of fish collected downstream of the spill site included alcohol and phenol Phase I metabolites associated with exposure to C_2 - C_4 substituted naphthalene, phenanthrene, fluorene and dibenzothiophene. These compounds were found to be present in the spilled condensate. The highest concentration of metabolites found in the bile of exposed fish were observed to be alcohols associated with exposure to C_2-substituted dibenzothiophene and fluorene. It is suggested that these metabolites be monitored to determine whether cleanup efforts are sufficient to reduce future exposure. Bile analyzed from fish taken upstream of the spill site did not contain any of the metabolites found in the exposed fish. This information was collected to determine the potential impact of the spill on the aquatic receiving environment.

049 The Effect of a Chemical Dispersant on the Bioavailability and Trophic Transfer of a Petroleum Hydrocarbon to Topsmelt. Mielbrecht, E.E.*, Wolfe, M.F., Schwartz, G.J.B., Singaram, S. and Tjeerdema, R.S., University of California, Santa Cruz, CA.; Sowby, M.L., California Department of Fish and Game, Sacramento, CA. The environmental implications of utilizing chemical dispersants in oil spill clean up are not well characterized. Application of chemical dispersants speed the dissolution of a surface oil slick by increasing the apparent solubility of spilled oil. What is poorly understood is how chemical dispersing agents alter the bioavailability and subsequent bioaccumulation of these dispersed hydrocarbons. The goal of this research was to elucidate the effects of a chemical dispersant on the bioaccumulation of hydrocarbons in a marine fish. Uptake, biotransformation and depuration of a model hydrocarbon, ^{14}C-phenanthrene, by larval Topsmelt, *Atherinops affinis,* via aqueous-only or combined dietary and aqueous routes of exposure were measured and compared for the water accommodated fractions of chemically dispersed and undispersed Prudhoe Bay Crude Oil. Dietary uptake was measured by providing pre-exposed *Branchionus plicatilis,* a marine rotifer, as a food source. Uptake of ^{14}C-phenanthrene was measured over twelve hour exposure periods followed by twelve hours of depuration in clean sea water. All compartments including whole organism tissue and exposure media were analyzed to quantify uptake or depuration and biotransformation of ^{14}C-phenanthrene using high pressure liquid co-chromatography and liquid scintillation counting. Results show that bioconcentration of phenanthrene is significantly decreased in fish when a simulated oil spill is treated with a dispersant ($p<0.05$). When trophic transfer is included, bioaccumulation of phenanthrene is delayed but eventually greater when the dispersant is used ($p<0.05$). These results suggest that the use of chemical dispersants in oil spill clean up will decrease the immediate bioconcentration of polycyclic aromatic hydrocarbons in fish, but the eventual biomagnification of these hydrocarbons will increase. This information will aid in decisions concerning the use of chemical dispersants in oil spill cleanup.

050 Brine Shrimp Assay-guided Isolation of Toxic Compounds from Sediments near Aluminum Processing Plants. Opinya Ekabo*, School of Public Health, SUNY, P. O. Box 509, Albany, NY 12201-0509, Patrick O'Keefe, Brigitte Bachner, Robin Storm, Wadsworth Center, New York State Department of Health, P. O. Box 509, Albany, NY 12201-0509. Polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs) and polychlorinated dibenzo-p-dioxins (PCDDs)/ polychlorinated dibenzofurans (PCDFs) are the four major classes of non-polar organic pollutants which have been identified in sediments collected from an aluminum processing plant near Massena, NY. It has been observed, in our laboratory, that sediments depleted of these contaminants by supercritical fluid extraction with CO_2 were still very toxic to larvae of the midge, *Chironomous tentans*. In our continued efforts to identify and isolate novel environmental pollutants around the Massena aluminum plant, we have employed activity-guided fractionation of sediment extracts using the brine shrimp (*Artemia salina*) assay in the identification of several oxidized PAHs and aromatic nitrogenous compounds in the sediments. The structures of six compounds: fluoren-9-one, carbazole, anthraquinone, 9-anthracenecarbonitrile, benzanthrone and benz(a)anthracene-7,12-dione have been confirmed unambiguously by comparing their GC-MS and GC-MS/FT-IR data with those of authentic samples. The estimated amount of the known compounds per dry weight of sediment is as follows: fluoren-9-one (0.18 ppm), carbazole (1.36 ppm), anthraquinone (2.96 ppm), 9-anthracenecarbonitrile (0.042 pm), benzanthrone (0.08 ppm) and benz(a)anthracene-7,12-dione (0.35 ppm). Fluoren-9-one was found to be very lethal to the shrimp, causing 100% mortality at the 3-ppm level. Carbazole, benzanthrone and benz(a)anthracene-7,12-dione exerted narcotic effects on the shrimp.

051 Environmental Pertubations and Coral Reef Ecosystems. Ostrander,G.K.*, The Johns Hopkins University, Baltimore, MD; E. Scully, Towson University, Towson, MD. The ongoing degradation of the world's coral reefs ecosystems has been well documented. Mass extinctions of invertebrates (e.g. sea urchins), over-fishing, outbreaks of coral diseases, and other stress- related events (e.g. coral bleaching) are a few of the events known to be occurring. Corals are the infrastructure of the reef community, yet no less than 10 different disease processes are occurring among hard and soft corals world-wide. Examples include white-band disease, black-band disease, and rapid wasting disease. Additionally, the frequency of occurrence and range of coral bleaching, in which corals loose their symbiotic algae, has steadily increased. Common threads linking these processes include their wide-spread global occurrence, rapid effects, and overall dramatic harm they cause to the reef ecosystem. Three lines of study have been initiated in our laboratories. Results of a ongoing long-term study at a remote coral reef in the Carribean will be presented. Ten permanent transects were established in 1994 and sampled at regular intervals through early 1998. Repeated Measures Analysis of Variance, in which the proportion of the total transect length occupied by each taxonomic category were compared, revealed significant temporal differences in species composition. Likewise, changes in fish community structure have been documented. Finally, we will report on our initial efforts in establishing coral cell cultures in the laboratory.

052 The Effects of Copper and Lead on the Fertilization Success and Larval Survival of the Scleractinian Coral *Goniastrea aspera*. Reichelt, A.J.*, Harrison, P.L., Centre for Coastal Management, Southern Cross University, Lismore, Australia. The objectives of this study was to collect toxicological information for copper and lead on the fertilization success and larval survival of *Goniastrea aspera*. EC50 and LC50 values could then be estimated for early life stages of corals and comparisons made with data on other marine organisms. The fertilization toxicity tests were done at sites on the Great Barrier Reef using gametes from spawned corals in 1994 and 1996. Groups of approximately 100 eggs were crossed with sperm after 30 minutes exposure to various concentrations of copper or lead (five replicated were used for each exposure). After a five hour development period the percentage fertilization was determined. EC50 and NOEC values were then estimated from the results. Two larval survival experiments were done for copper and for lead using larvae grown from fertilized gametes from coral spawning in 1995. Twenty healthy larvae were placed in vials with various concentrations of copper or lead (five replicated were used for each exposure). The larval survival was monitored at set time intervals from the dose time. EC50 and NOEC values were then estimated from the results. EC50 values for the effect of copper on coral fertilization success were 14.93 mg/L and 18.52 mg/L for tests done in 1994 and 1996 respectively. The EC50 value for lead was 2467 mg/L for an experiment done in 1996. The NOEC values for copper were 2 mg/L and 12.8 mg/L for tests done in 1994 and 1996 respectively and were 5455 mg/L for lead. The 48 and 72 hour LC50 values for the effects of copper on larval survival were 39.84 mg/L and 34.08 mg/L for the first experiment and 87.31 mg/L and 81.62 mg/L for the second experiment. The 48 and 72 hour LC50 values for lead could not be estimated for the first experiment and was 9890 mg/L for both times in the second experiment. The 48 and 72 hour NOEC values for copper were 20 mg/L and 5 mg/L for the first experiment and 50mg/L for both times in the second experiment. The 48 hour NOEC for the effects of lead on larval survival was 5000 mg/L. It is clear that fertilization success is more sensitive to copper and lead compared to larval survival. Fertilization success is essential for the long term maintenance or reef coral populations. However, corals remain in the larval phase for longer periods of time and may encounter pollutants at any time, and be exposed for longer periods during this motile planktonic phase.

053 Understanding Risks to Tropical Marine Ecosystems from Oil Spills. Gardiner, W.[1*], Neff, J[2], and Stejskal, I[3]. [1]Battelle Marine Sciences Laboratory, Sequim WA; [2]Battelle Ocean Sciences, Duxbury, MA; [3]Apache Energy Corporation, Perth, Australia. To better understand the potential risk of oil spills to tropical marine ecosystems, a suite of toxicity tests using tropical marine species (clownfish, white shrimp, coral, echinoderm larvae) was developed and conducted with the water-accomodated fractions (WAF) of six crude oils and their weathered fractions. Chemical and physical properties of each of the crude oil, weathered fraction, and the WAF were also analyzed. Crude oils were collected from the Northwest Shelf of Australia and included condensate, light-, medium-, and heavy-weight oils. Oils were artificially weathered by evaporative topping to simulate 1-3 hours, 0.5-1 day, and 1 week of weathering. After 1 week of weathering, the condensate and light crude had almost completely evaporated. Approximately 54% and 18% volume of the medium and heavy crudes were lost after 1 week. Toxicity was highest in WAF prepared from the unweathered condensate and light-weight crude; however, toxicity was almost completely removed after 0.5-1 day weathering. Virtually no toxicity was associated with the WAF prepared from the heavy crude or its weathered fractions. WAF prepared from the medium-weight crude had relatively moderate toxicity; however, toxicity remained after one week of weathering. The results of the chemical analyses indicate that toxicity in the light fractions was primarily due to volatile alkanes and monocyclic aromatic hydrocarbons. After 1 day of weathering, light alkanes and MAHs were lost. The remaining toxicity (as in the medium crude) was caused primarily by PAHs and phenols. This study represents the first attempt to relate the changes in physical properties of crude oils during the evaporative weathering to changes in their chemical composition and toxicity to marine organisms.

054 Effects of Nutrient Enrichment on Caribbean Corals: Laboratory Experiments vs Field Validation. Davies, P.S.*, Marubini, F., Institute of Biomedical & Life Sciences, University of Glasgow, U.K. Preparations of the corals *Porites porites* and *Montastrea annularis* in Barbados were maintained in a laboratory photostat in control oligotrophic seawater and in elevated levels of nitrate and phosphate for three weeks. After exposure to high nitrate (5mM and 20mM), the corals had a higher rate of photosynthesis per surface area, and a higher population density of zooxanthellae. The freshly isolated zooxanthellae also had higher levels of chlorophyll and protein. Conversely, in high nitrate, the rate of skeletogenesis decreased. Exposure to high phosphate (up to 5mM) resulted in no change in productivity but the rate of skeletogenesis was reduced in *P.porites* only. Attempts were then made to validate these results by exposing the corals for a similar period at three sites along a eutrophication gradient in the sea. It was found that the results were confounded by another environmental variable, in the form of turbidity and light availability. Discriminate function analysis separated those physiological variables that are most sensitive to nutrient enrichment (primary characters) and from those that are highly dependent on light levels and only secondarily on nutrient levels. Chlorophyll concentration emerged as a primary character and therefore the most sensitive indicator of nutrient enrichment. By contrast, rates of gross photosynthesis, respiration rate and rate of skeletogenesis were secondary characters and therefore poor discriminators in the field, in situations where light levels varied independently of nutrients. The roles of laboratory-based and field-based bioasssays will be discussed in the light of these observations.

055 The Effects of Coastal Pollution on Reproduction and Recruitment of Tropical Marine Benthic Invertebrates. Richmond, R.H.*, Leota, S., University of Guam, Mangilao, Guam; Crosby, D.G., University of California at Davis, Davis CA. The majority of coral reef benthic invertebrates reproduce by spawning their gametes into the water column, where larval development and dispersal occur. Subsequently, competent larvae must find appropriate substrata upon which to settle and metamorphose. Studies of mass-spawning scleractinian corals have identified five chemically sensitive links responsible for successful reproduction and recruitment. These include reproductive synchronization among conspecifics, egg-sperm interactions, embryological development, metamorphic induction and acquisition of symbiotic algae. Coastal pollution from runoff has been demonstrated to result in reproductive failure in corals. Fresh water dilution, from 35 ppt to 28 ppt, alone may result in an 80% drop in fertilization rates. The presence of lateritic soil in coastal waters causes a similar drop in fertilization rates corresponding to a much smaller drop in salinity. The organophosphate pesticide chlorpyrifos has been found to inhibit settlement and metamorphosis of healthy coral larvae at levels of ppb. Reduced recruitment rates were observed in experiments where either larvae or substrata were exposed. These experiments demonstrate differential effects of pollutants on different life-history stages of invertebrates as well as differences in the way hydrophilic and hydrophobic substances affect reproduction and recruitment. The studies also show that standard protocols, including LC- 50's may be non-predictive and of limited use in protecting coral reef ecosystems.

056 The Occurrence of Developmental Defects in Embryos of the Damselfish, *Abudefduf sordidus* (Pomacentridae), Relative to Polychlorinated Biphenyl (Pcb) Contamination at Johnston Atoll, Central Pacific Ocean. Kerr, L. M., University of Massachusetts, Boston, MA. The occurrence of developmental abnormalities in the embryos of a demersal spawning fish was used to determine if adverse effects due to anthropogenic contamination could be detected at Johnston Atoll. *Abudefduf sordidus* embryos were taken from four spawning colonies that occurred in areas with differing polychlorinated biphenyl (PCB) sediment concentrations. Of the three colonies occurring off of Sand Island, two were in areas with intermediate and high PCB concentrations (both exceeding ecological screening levels) while one was in an area with low PCB concentration. The control colony was 1000 m from Sand Island and occurred in an area with low sediment PCB concentrations. Early stage embryos were examined for abnormalities defined as deviations from normal developmental stage or morphological differentiation. A sample consisted of 200 embryos taken from a newly spawned clutch. The mean number of abnormalities in clutches from the control colony (1.4%, N = 71) was significantly different from the mean number of abnormalities at the site with the highest PCB concentration (9.3%, N = 53); (Kruskal Wallis P = 0.001). In addition, the control site was significantly different from all Sand Island sites combined (5.6%, N = 145); (t-test P < 0.001). Linear regression analysis indicated a significant relationship between the mean occurrence of abnormalities and mean sediment PCB concentration ($r^2 = 0.86$, P = 0.048). Reproductive output, in terms of number and size of clutches spawned, was not significantly different among colonies. These results suggest that while embryonic abnormalities were elevated at the high PCB concentration area, adult reproductive processes were not affected.

057 High Volume Discharges of Oil Well Produced Water to the West Java Sea, Indonesia: Chemical Composition and Environmental Effects. Neff, J.M.*, Battelle Ocean Sciences, Duxbury, MA; Smith, J.P., Maxus Southeast Sumatra, Inc., Jakarta, Indonesia. Large volumes of produced water are pumped to the surface with oil and gas on production platforms. Three production processing platforms operated by Maxus Southeast Sumatra in the West Java Sea generate between 17.3 and 141 million liters/day of produced water which is treated to remove petroleum and discharged to the sea. The produced waters contain several metals, the most abundant being barium (to 276 mg/L), zinc (to 16.7 mg/L) and arsenic (to 8.5 mg/L). Only traces of radium isotopes are present. The metals are not present at concentrations high enough to cause injury to water column organisms following initial mixing of produced water in ambient sea water. The produced waters also contain 331 - 3638 mg/L BTEX, 231 - 745 mg/L total PAHs, and 2490 - 6329 mg/L total phenols. The produced waters have an acute toxicity to mysids of 18.4 - 34.8 volume percent produced water and to silverside minnows of 12.3 - 19.5 percent. Modeling showed that initial dilution of the produced water following discharge to the ocean ranged from 12-fold to more than 100-fold, depending on discharge rate and local oceanographic conditions. A risk assessment showed that local zooplankton, but probably not pelagic fish, may be adversely affected if they are entrained in the diluting produced water plume,

058 Weathering of Hydrocarbons in Mangrove Sediments: Testing the Effects of Using Dispersants to Treat Oil Spills. Burns, K.A., Codi, S.*, Pratt, C. and Duke, N. AIMS, Queensland, Australia. This field study was a combined chemical and biological investigation of the relative effects of using dispersants to treat oil spills impacting mangrove habitats. The aim of the chemistry work presented here was to determine whether dispersants affected the rate of penetration, dissipation or long term retention of a medium range crude oil (Gippsland) stranded in a tropical mangrove environment. Permission for a planned oil spill was granted in the Port Authority area of Gladstone, Queensland (Australia). Sediment cores from three replicate plots of each treatment (oil only and oil plus dispersant) were analyzed for total hydrocarbons and for individual molecular markers (alkanes, aromatics, triterpanes). Sediments were collected at 2 days, then 3, 6 and 12 months post-spill. Over this time oil decreased exponentially from 47 mg/g dry wt to 5 mg/g dry wt (n=4). There was no statistical difference in initial oil concentrations, penetration of oil to depth, or in the rates of oil dissipation between oiled or dispersed oil plots. Dispersant use did not alter the triterpane signature of the retained oil, which is important for litigation purposes. The predominant removal processes were evaporation and dissolution, with a lag-phase of 1 month before the start of microbial degradation. Since dissipation of the oil was not related to pre-treatment with dispersant, other relevant sediment factors were considered. The most residual fraction of the oil that remained after 6 months, the higher molecular weight hydrocarbons, correlated with the TOC content of the soil. The chemistry data also provided the context for interpretation of the biological observations (including mangrove physiology, geomorphology, and burrowing animal populations).

059 Seasonal Water Quality Study of Riverine and Coastal Waters Effecting the Coral Reefs of Veracruz, Mexico. Ricono, N.A. M.S. Thesis, Texas A&M University-Corpus Christi. Physical and Chemical water quality parameters were studied to identify seasonal variability in coastal and terrestrial waters thought to be related to the decline in growth and distribution of coral populations in the southwestern Gulf of Mexico. Over 20 emergent, platform-type coral reefs exist along the southern portion of the state of Veracruz, Mexico. These reefs are of interest ecologically and geologically as they are influenced by considerable pluvial input and are located within a terrigenous sedimentary province. This has led to the depauperate nature of these reefs, containing a maximum of 28 hermatipic coral species. Prior to the 1970's, these reefs were described as containing 50-100% cover of *Acropora palmata* and *A. cervicornis* in the shallow reef zones and 40-60% cover of *Montastrea annularis* in deeper reef zones. Since the early 1970's there has been a steady decline in coral distribution leading to 1-5% cover "A. palmata" and "A. cervicornis" and 76-82% algal cover. Possible causes of this rapid coral decline include seasonal increases of riverine discharge containing large sediment loads, nutrients, and pesticides from agricultural, industrial, and municipal runoff. Seasonal changes in chemical and physical water quality parameters were tested by taking pre-rainy season, rainy season, and post-rainy season samples from riverine and coastal waters. Results indicated no significant difference between seasonal nutrient concentration, salinity, or suspended solids. The freshwater inflow events reported to be common during the rainy season were not observed during this survey. Their influence, however is substantial in shallow water environments leading to the lack of coral cover in that area. The survival of large boulder coral species at deeper depths relate decreases in coral cover to high sedimentation rather than high nutrient concentrations.

060 Investigation of Unknown Chemical Contamination on Laysan Island, Hawaiian Islands National Wildlife Refuge. Woodward, L.A.*, USFWS, Honolulu, HI; Scow, K.M., University of California-Davis; Palawski, D.U., USFWS, Honolulu, HI. The Dead Zone is an approximate $10m^2$ area on the northern edge of Laysan Island of the Hawaiian Archipelago. It was first noted as being unusual approximately 10 years ago by biologists who found a large number of Laysan albatross (*Diomedea immutabilis*) carcasses at the site. On closer inspection it turned out the carcasses were not being consumed by crabs, and in fact, decomposers such as crabs and flies were found lying dead both on the dead birds and on the ground not in the immediate vicinity of the carcasses. Live and seemingly healthy albatross chicks and adults were also present within the zone, suggesting that the birds were not affected by what was killing the crabs and flies. The results of chemical analyses ruled out that chemical warfare agents and numerous pesticides and other substances were present in the Dead Zone sand. The one substance that has been found in some, but not all samples, is carbofuran. The symptoms exhibited by dying flies and crabs are consistent with exposure to carbofuran. According to available literature, in sandy soil at pH 8 (Laysan sand pH), carbofuran should degrade in a few months. However, while at an apparent reduced level, the Dead Zone is still killing invertebrates after 10 years.

061 Technical Problems Encountered by Restoration Projects: a Failure to Learn from Our Mistakes. Willard,D.E. and Oster, C.V.,Jr., School of Public and Environmental Affairs, Indiana University, Bloomington, Indiana. We divide restoration problems into three interrelated categories: Technical limits, regulatory, legal and institutional angles, and social and economic tradeoffs and consequences. Engineers and ecologists working together can improve the ecological functions of many ecosystems. Failures result for two reasons: a lack of attention to design and follow through, and a lack of agreement on the definition of success. Careful site selection and fitting of projects can cure the first failing, by fitting site design to landscapes, then, following construction with regular monitoring and post construction management. The subjective view of the definer heavily predetermines the second sort of failure. Usually conflict exists between definitions based on pre-Columbian conditions of the ecosystem and the practical limits imposed by current conditions. Laws, regulations and the institutions that support them have a difficult role in that the functions, values and processes crucial to predetermining the success of an ecosystem restoration project are necessarily adaptive and temporally changing. While, these institutions wish to have rules that they can equitably apply to all. However, each situation varies technically and socioeconomically. Therefore, a need exists for precise definitions for imprecise things. This lack results in the even application of illfitting rules to all cases (the one size fits none rule.) For example few jurisdictions differentiate between restoration for restoration's sake and restoration to fulfil some permit obligation. Follow-up monitoring and enforcement are often lacking.

062 Restoration of Seagrasses in Tampa Bay Florida: What Have We Learned and What Do We Need to Know? Bell,S.S. University of South Florida, Tampa, FL. In seagrass restoration studies in Tampa Bay, we have monitored multiple sites over 2-7 years. Our research has focused on assessing faunal responses to seagrass introduction and seagrass repopulation. The restoration of seagrass has been successful. Seagrass vegetation has been established and expanded. Likewise, faunal abundances in restoration sites mirror those from natural areas within a two-year period after seagrass introduction. Specifically, abundance, size distribution and species composition of small fish, residents in seagrass beds, were not distinguishable from that of natural sites after 17-22 months after restoration began. Our studies indicate that this pattern persisted for 7 years. Further, examination of trophic links has shown that fish feed similarly in restored and natural areas within 2 years after seagrass is introduced. Thus, faunal functional equivalency, appears to occur within a short time after seagrass planting, even when seagrass density has not reached natural levels. What remains to be tested is whether these restored sites maintain similar functional equivalency over longer (decades) time intervals and how they respond to disturbance. We also have information on other fauna in restored sites. In restored plots we found that, in some cases, polychaete productivity can be enhanced compared to natural areas. This work has spawned concern about the choice of appropriate "control" areas in evaluating restoration. Recent studies have suggested that a landscape approach to restoration may be useful for shallow water systems and the results obtained from our earlier work may be related to site position within the estuary and/or characteristics of adjacent vegetation patches. New efforts will be required to determine if a landscape approach, which considers spatial arrangement of restored and natural sites, will be useful for designing and evaluating seagrass restoration.

063 Macroscale Restoration of Coastal Wetlands in Disturbed Environments. Weinstein, M.P., New Jersey Marine Sciences Consortium, Sandy Hook Field Station, Fort Hancock, N.J., and Wainright, S.C., Institute of Marine and Coastal Sciences, Cook College Rutgers University, New Brunswick, N.J. Too often, the goals of tidal marsh restoration are tied to unrealistic endpoints. Although general consensus reached among stakeholders on how to proceed, "reference" marshes are frequently chosen for their relatively undisturbed condition, setting an unrealistic upper-bound for restoration success. On the other hand, the large uncertainties and lack of practical tools for measuring marsh function tend to over-simplify the success criteria established by regulatory agencies. Case histories are shown where previous disturbance history of restored sites leads to a wide-range of hydrogeomorphic (physiographic) endpoints while at the same time satisfying the permitted criteria for vegetation cover and percent open-water. We hardly know how natural marshes function, how do we apply this limited knowledge to reconcile the vastly different configurations of restored marshes that are ecologically engineered? It is not surprising that the complexity of natural marsh functions, and our difficulty in measuring them, makes it exceedingly difficult to predict the outcome of *any* restoration project. More careful planning and development of restoration goals is needed up front for the process to work.

064 Large Scale Regional Restoration Planning Successes and Lessons Learned: Pacific Northwest Case Studies. Clark, Jr., R.C., NOAA Restoration Center Northwest, Seattle, Washington. Six major natural resource damage assessment (NRDA) settlements have been reached in Elliott and Commencement Bays, Puget Sound, Washington, to compensate for injuries to trust fish, wildlife, and habitat resources from the release of hazardous materials under the auspices of Superfund (CERCLA). These six settlements were reached over an 8-year period and contained different sets of negotiated conditions and parameters for accomplishing the desired restoration goals. Baywide (regional) restoration goals were established and integrated into, or formed a basis for later, larger watershed restoration plans. Several habitat restoration projects have been completed while others are under construction or being planned and long term monitoring/steward efforts underway. The challenge for the regional Natural Resource Trustees (Federal, state, and tribal) has been to integrate numerous specific projects into a landscape ecology approach covering an entire river watershed (e.g., Duwamish/Green, Puyallup/White) with primary emphasis on restoring damaged habitats in dynamically-changing, highly industrialized, urban estuaries (e.g., Seattle's Elliott Bay, Tacoma's Co. and Bay). The guiding region document in Elliott Bay is the Elliott Bay/Duwamish Restoration Program's Concept Document based in large part on a very detailed U.S. District Court Consent Decree signed with the City of Seattle and King County/METRO. The settlements covered by five individual and differing Consent Decrees in Commencement Bay are guided by a 1997 Programmatic Environmental Impact Statement and Restoration Man and implemented under a NRDA Trustees Council and Restoration Case Manager. The variety of approaches and criteria provide an excellent opportunity to compare and contrast the strong and weak points of each settlement and their effect on restoring aquatic habitats in the Pacific Northwest.

065 From Controversy to Consensus: The Houston-Galveston Navigation Channels, Texas Project. Bass, R.J., U.S. Corps of Engineers, Galveston District, Galveston, Texas. The Houston-Galveston Navigation Channels, Texas project is the largest dredging project ever undertaken in Texas. The Houston Ship channel (HSC) extends about 60 miles across Galveston Bay, Texas' most productive estuarine system, from the Gulf of Mexico to the Port of Houston, the Nation's second largest port. Dredging will take a minimum of four years, excavate about 350 million cubic yards of material, and cost about 560 million dollars to complete. Planning of this large project has had a long and controversial history. The Galveston District's early efforts began in 1967 and ended with completion of the Galveston Bay area Navigation Study in 1987. This report recommended widening and deepening the channel and disposing of 400 million cubic yards of dredged material unconfined over about 11,000 acres of bay bottom. No beneficial uses of the dredged material for wetland restoration were recommended. The report and its associated Final Environmental Impact Statement met with almost complete condemnation from Federal and State resource agencies, the environmental community, and general public. At this point, the project was on the verge of referral to the Council On Environmental Quality. Had it past that process, it would have likely been litigated for years. Today, after redesigning the project to include wetland restoration and completing a Supplemental

Environmental Impact Statement in 1995, the project is under construction. This was achieved by formation of an Interagency Coordination Team (ICT). This was a long, difficult and expensive undertaking. This presentation will discuss the role of the ICT in regional project planning to use all dredged material beneficially to restore fish and wildlife habitat through the bay system and how the Galveston District guided the project from controversy and chaos to consensus and construction.

066 Regional Restoration Planning in the New York/New Jersey Harbor Estuary: Problems and Approaches. Catena, J.G. NOAA Restoration Center, Gloucester, MA. Several significant oil spills in New York Harbor in the early 1990's led to multimillion dollar settlements paid to state and federal governments by responsible parties for injuries to natural resources, which resulted in funding for the development of a regional restoration plan. The regional plan identifies the following types of "restoration" priorities for the region: (1) restoration and enhancement of wetlands, shellfish, finfish, wading birds, and waterfowl; (2) acquisition of wetlands and adjacent upland buffers in danger of being developed; (3) enhancement of public access to the harbor; and (4) public education. Since the development of that plan, other organizations have also identified regional restoration priorities for both the harbor as a whole and for specific watersheds or embayments. The several regional restoration planning efforts in the New York/New Jersey Harbor area have: (1) created a core group of agencies, academics, and environmental organizations dedicated to the oversight and implementation of restoration projects in the region; (2) created a working menu of restoration options to choose from as funding becomes available; and (3) focused those dollars on the highest priority projects. However, regional restoration planning and implementation has had to overcome several obstacles: (1) initial disagreements by some agencies to spend money on planning rather than in-the-ground restoration: (2) jurisdictional squabbles (City vs. State, and State vs. State) over how to spend restoration dollars — brought about by differing attitudes about the relative importance of the priorities established in the regional plan; (3) a scattershot approach to planning rather than a true ecosystem-based approach to determining highest priority problems and solutions in the region; and (4) the realization of the data intensive nature of a regional planning effort.

067 The Regulatory Evolution of Natural Resource Damages. Case Studies. Brody, N.S. Atlantic Richfield Company, Los Angeles, California. Following nearly a decade of rule proposals and promulgations, the Department of Interior (DOI) enacted a final comprehensive Natural Resource Damage Assessment (NRDA) Regulation under Superfund on March 25, 1994. The focus of the rule is to assess natural resource damages through a restoration and compensation determination plan. Subsequently, on January 6, 1996, the National Oceanic and Atmospheric Administration (NOAA) promulgated its NRDA rule under the Oil Pollution Act (OPA). The NOAA rule dramatically alters the focus of the NRDA, or at least the terminology of the focus, from the DOI rule. Under OPA, instead of assessing restoration damages and compensable value damages, trustees are directed to measure primary restoration and compensatory restoration measures. Primary restoration is defined to include restoration of resources to baseline, or pre-incident condition. By definition, compensatory restoration is supplemental restoration actions beyond those necessary to achieve actual restoration. Finally, on July 16, 1996, as part of a Biennial review of its NRDA regulation, DOI solicited comment on whether it should consider adopting NOAA's "new method" for measuring natural resource damages. Through the use of case-specific examples the presentation will discuss the practical impacts of these changes including, in particular, their positive and negative attributes.

068 Using Partnerships to Solve Natural Resource Restoration Problems in Two Major Urbanized Puget Sound Estuaries. Clark, Jr., R. C., NOAA Restoration Center Northwest, Seattle, Washington. Natural resource restoration can provide real-time solutions to reversing environmental degradation of urban aquatic habitats using funds recovered from responsible parties (polluters). For habitat restoration to be successful, the process requires a close partnership and a commitment between natural resource trustees, other resource agencies and land managers, and the public. In most cases, the results can be significantly enhanced by the active and voluntary involvement of the responsible parties. Major natural resource restoration efforts are in progress in two large, commercially-important, and contaminated urban estuaries in the Pacific Northwest's Puget Sound, Washington. The myriad of problems encountered in Elliott Bay (Seattle's harbor) and the Lower Duwamish River with its initial $24 Million habitat development (5 projects) and sediment remediation (4 projects) are compared and contrasted with $22 Million natural resources damage settlements (CERCLA) in Commencement Bay (Tacoma's harbor) and the Puyallup River watershed. In the latter, a bay-wide programmatic Environmental Impact Statement and Restoration Plan has been completed using an ecosystem or landscape ecology approach. A pilot project has been completed to test restorative processes by converting an industrial upland fill to productive upper intertidal habitat. Through a management team approach spearheaded by the federal, state and tribal natural resource trustees in partnership with other resource and regulatory agencies, local municipal governments, user groups, environmental interests, the public, and responsible parties, some major strides have been made in solving many of the problems of practical habitat restoration. Consensus processes have been developed to attack most of the remaining areas of disagreement so that actual improvement of natural resources and their habitats can take place with a mini mum of delay and transaction costs.

069 Restoration Partnerships: A Win, Win, Win, Win Proposition. Campbell, T.A. and Strong, A. L. Campbell and George, L.L.P., Houston, Texas. Making the best of a bad situation is the lot of a company that has an unpermitted discharge of a listed contaminant or the discharge of oil. Dealing with clean-up issues has been complex, but companies, NGOs and government agencies have settled into well-understood norms. These norms, while contentious, are accepted. In initial Natural Resource Damage Assessments, PRPs, Government Agencies and Environmental NGOs naturally followed the patterns laid down in the clean-up arena. However, as the focus in the NRDA arena has shifted (in portions of the country) from monetary damage claims to restoration-based compensation, those relationships have shifted and the potential for win, win resolutions have become a possibility. Some Environmental NGOs and governmental agencies unwittingly benefitted from these changing relationships, while others have positioned themselves to benefit from this new paradigm. This presentation will focus on describing how restoration-based compensation has benefited NGOs, PRPs, Government Agencies and the Public, thereby creating Win, Win, Win, Win situations. It will use case studies from oil and chemical spills where restoration-based compensation was used and all parties were benefitted. The manner in which they were benefitted will be discussed. From these examples, we will move to specific steps that the parties can take to position themselves in advance to take advantage of the benefits of restoration-based compensation.

070 On the Evolution of an Oil Spill Restoration Plan: Facts and Fiction of Restoration and Monitoring of an Urban Intertidal Salt Marsh. Matsil, M.A., Alderson, C., Bergen, A., City of New York, Parks & Recreation, New York, New York. In 1990, one million gallons of oil spilled into the Arthur Kill, a narrow body of water separating New York City and New Jersey, severely damaging intertidal salt marsh and associated wildlife communities. After protracted negotiations, a settlement led to the funding of an experimental salt marsh restoration of heavily oiled substrate. Bioremediation has resulted in reduced petroleum hydrocarbons and accelerated erosion rates, and initial recovery of the salt marsh ecosystem. The project has focused on establishing comprehensive monitoring protocols in restored and unrestored sites and elucidating a relationship between heterotrophic bacteria capable of degrading petroleum hydrocarbons found in the rhizosphere of the restored Spartina altemiflora plants, and the reductions of the Total Petroleum Hydrocarbons (TPH). TPH ranged from 160 to 57,000 ppm. To date, more than 500,000 Spartina alterniflora plants have been restored, propagated from indigenous seed to areas heavily impacted by the oil with the assistance of 500 volunteers. Restored and unrestored control sites have been established; monitoring includes bacterial analysis, fertilizer study, plant productivity, invertebrate population study, fish, avian and mammal breeding studies in 120 M2 quadrant. Unrestored quadrant in oil impacted reference sites have resulted in no voluntary seed or plant recruitment, and remains denuded and subject to increased erosion eight years after the spill.

071 Response of Aquatic Communities to a Model Alcohol Ethoxylate and an Ecosystem Level SAR. Belanger, S. E.*, Guckert, J. B., Bowling, J. W. and D'Angelo, D. J. The Procter & Gamble Company, Cincinnati, Ohio USA. The P&G Experimental Stream Facility (ESF) was used to evaluate a model, non-commercial alcohol ethoxylate (AE) with an average alkyl chain length of 13.5 and ethoxylate chain length of 5.7 (25-6 AE). Two experiments at the ESF were performed: AE added in river water at 0, 13, 36, 76, 259, and 760 mg/L and a second experiment in which AE was added at 0 and 37 mg/L with 0, 5, and 13% wastewater treatment plant effluent. Both experiments assessed responses of periphytic, protozoan and macroinverterbate communities to 25-6 AE for 8 weeks following 8 weeks of colonization (16 weeks total). Supporting acute and chronic toxicity studies (daphnids, clams, snails) were also conducted. A broad spectrum of effects were observed for AE in river water. Periphytic communities were enhanced (biomass, activity) at high AE concentrations as a result of indirect interactions between AE and river water solids. Invertebrate communities were adversely affected (mayflies, caddisflies, selected molluscs) at >76 mg/L by 4 weeks and >13 mg/L by 8 weeks. Both effluent and AE impacted aquatic communities although effluent-related effects were of a much greater magnitude. When results were combined with several other AE model ecosystem studies conducted by others between 1992-1996, a reasonable structure-activity-relationship (SAR) emerged. The SAR suggested 25-6 AE was more toxic than expected and a more hydrophobic AE (45-7) was less toxic than expected. Chronic toxicity tests for 25-6 AE resulted in NOECs ranging from 80 mg/L (*Daphnia magna*) to 760 mg/L (snails). Single species, model ecosystem and current environmental monitoring data indicate AE poses minimal risk to the environment.

072 Effects Assessment of an Alcohol Ethoxylate Using Outdoor Artificial Streams. Tattersfield, L.J.*, Young, L.J., Davies, E.H., McCarthy, W.V. Shell Research and Technology Centre, Thornton, UK. The use of surfactant compounds in detergents typically results in discharge "down the drain" and subsequent treatment in sewage treatment works. alcohol ethoxylates (AEs) as a class are large volume, nonionic, surfactant molecules used mainly in laundry and cleaning products. Linear-type AEs are readily biodegradable and the majority is removed via waste water treatment. Previous monitoring studies of AEs in effluent from representative municipal sewage treatment plants in the Netherlands have shown that the composition discharged had an average alkyl chain length of 13.3 and an average ethylene oxide number of 8.2. Generation of comprehensive effects data for an AE of a composition relevant to the average AE discharged from sewage treatment plants could significantly contribute to the strengthening of the risk assessment for AEs entering the environment. A commercial linear-type C12-15 AE9 compound was selected for testing as it has an average composition close to that determined in the monitoring study. Comprehensive effects data was generated using a system of 8 outdoor artificial streams. Mean measured exposure concentrations during a 56 day exposure period were 20, 40, 70, 160, 300, 390 and 740 mg/l plus an untreated control with doses randomly allocated to streams. Multivariate community-level analysis of invertebrates indicated a dose-response effect with communities becoming progressively dissimilar with increasing AE concentration. Individual and population level NOECs were in the range 70 to >740 mg/l. Invertebrates and fish were of similar susceptibility with *Simulium sp.*, *G. pulex* and *Baetis sp.* (population density) and *O.mykiss* (growth) being most susceptible. Algae were less sensitive and did not appear susceptible to the AE tested, most observed changes being secondary effects due to reduced grazing pressure.

073 Bioconcentration and Biotransformation of the Nonionic Surfactant Octaethylene Glycol Monotridecyl Ether $^{14}C\text{-}C_{13}EO_8$. Tolls, J.*, RITOX, Utrecht, NL; Sijm, D.T.H.M., RIVM, Bilthoven, NL. Alcohol ethoxylates (AE) are the most important nonionic surfactants used in household detergents. In this study we investigated the bioconcentration behavior of $^{14}C\text{-}C_{13}EO_8$ as a representative of AE in fathead minnow (*Pimephales promelas*). The rate constants of uptake and elimination of the parent were 323 $LHkg^{-1}Hd^{-1}$ and 10.1 d^{-1}, respectively. The parent compound specific bioconcentration factor (BCF) was found to be 32 $LHkg^{-1}$. The TLC chromatograms demonstrated that the $^{14}C\text{-}C_{13}EO_8$ derived radioactivity is incorporated into a large variety of chemical species. The large discrepancy between the BCF of the parent $^{14}C\text{-}C_{13}EO_8$ and the radiolabel is a qualitative indicator of the extent of biotransformation. The first-order one compartment model of bioconcentration was extended to allow for quantification of the *in vivo* rate constant of biotransformation of $^{14}C\text{-}C_{13}EO_8$ and the value obtained amounted to 10 d^{-1}. The model results indicated that biotransformation is the dominating contributor to elimination of AE. A comparison of $C_{13}EO_8$ to compounds of other classes of environmental relevance highlights the importance of biotransformation as process contributing to reduction of the bioconcentration potential of these compounds.

074 the Distribution of Degradation Products of Alkylphenol Ethoxylates near Sewage Treatment Plants in the Lower Great Lakes. Bennett, E.R.*, Trent University, Peterborough, ON, Canada; Metcalfe, C.D., Trent University, Peterborough, ON, Canada. Degradation of alkylphenol ethoxylate (APE) surfactants in the environment leads to the formation of relatively hydrophobic compounds such as nonylphenol (NP), octylphenol (OP), nonylphenol monoethoxylate (NP1EO) and diethoxylate (NP2EO) that have been shown to have estrogenic activity. In the first component of this study, sediment samples were collected from several sites (n=30) near industrialized and pristine areas of Lake Huron, Lake Erie and Lake Ontario and analysed for concentrations of NP and OP. These data indicated that NP was present at mg/g (dry weight) levels in sediments collected near urban and industrialized areas, and concentrations were highest near discharges from sewage treatment plants (STPs). In light of this, we collected sediment samples and deployed semi-permeable membrane devices (SPMDs) and caged freshwater mussels *(Elliptio complanata)* at several sites in the vicinity of an STP in Hamilton Harbour and two STPs in the Detroit River in order to determine the distribution of these compounds in sediment and water and their potential to bioaccumulate in aquatic organisms. NP, OP, NP1EO and NP2EO were found at mg/g (dry weight) concentrations in sediments and accumulated to ng/g (wet weight) concentrations in caged mussels near the STPs. However, in the Detroit River, the concentrations of these compounds declined to near background levels in the sediments, water column (i.e. SPMDs) and biota at stations approximately 1 km downstream from STPs. At stations in Hamilton Harbour, concentrations of APE degradation products also declined markedly in sediments, mussels and SPMDs located at stations a few hundred metres from the STP. These data indicate that the primary degradation products of APEs are present at ppm concentrations near wastewater outflows but their distribution is primarily confined to the immediate vicinity of these discharges.

075 Biodegradation of ^{14}C Ring-labeled Nonylphenol Ethoxylate in Activated Sludge and in River Water. Naylor, C.G.*, Huntsman Corporation, Austin, TX; J.B. Williams and P.T. Varineau, Union Carbide Corporation, South Charleston, WV; R.P. Yunick, Schenectady International, Schenectady, NY; K. Serak and C. Cady, ABC Labs, Columbia, MO; D.J. Severn, Jellinek, Schwartz & Connolly, Inc., Arlington, VA. The aerobic biodegradation of a ^{14}C ring-labeled nonylphenol 9-mole ethoxylate (^{14}C-NPE9) was examined in laboratory semi-continuous activated sludge (SCAS) and river water environments. The ^{14}C-NP used to prepare ^{14}C-NPE was more highly branched than the NP of commerce. In the SCAS experiments primary effluent from a local wastewater treatment plant was dosed with ^{14}C NPE9 and fed into triplicate SCAS systems, and the levels of ^{14}C in the clarified effluent, settled sludge solids, and CO_2 were monitored. The ^{14}C species in the effluent and sludge were partially characterized using liquid-liquid extraction and HPLC. A significant portion of the ^{14}C consisted of soluble metabolites that had degraded beyond the phenol ring. Dosing of the SCAS system ended after 28 days, and dissipation of the residual radioactivity was followed for another 19 days. CO_2 evolution and decline of radioactivity in the sludge solids both followed first order rate kinetics, with half-lives of 2.8 days and 5.8 days, respectively. A portion of the residual sludge activity was incorporated into the biomass. In the river die-away experiment, the extent of $^{14}CO_2$ evolution from river water dosed with ^{14}C NPE was monitored for 128 days. After an induction period of 21 days $^{14}CO_2$ evolution followed first order kinetics; the half-life was 22 days. 40% of the NPE aromatic ring carbon was converted to CO_2. This is the first unequivocal demonstration that the phenolic ring of NPE is mineralized under activated sludge and die-away conditions.

76a Statistical Risk Assessment of Nonylphenol and Its Ethoxylates In U.S. Rivers. J. A. Weeks*, Racine, WI; C. G. Naylor, Huntsman Corporation, Austin, TX; C. A. Staples, Assessment Technologies, Inc., Fairfax, VA; and J. B. Williams, Union Carbide Corporation, South Charleston, WV. Nonylphenol has been subjected to increasing research and scrutiny for two decades. Nonylphenol can be a degradation intermediate of nonylphenol ethoxylates, a major surfactant class. Various ecological effects, including disruption of endocrine systems, have been hypothesized to be the result of environmental NP concentrations. Sufficient information is available for US rivers to examine the potential for such risks using a comprehensive statistical approach. Data to support the risk assessment were drawn from recent reviews of the aquatic toxicity of alkylphenol ethoxylates, and the extensive research program of the Alkylphenols and Ethoxylates Panel of the CMA. Environmental concentrations were based on a survey, planned in cooperation with EPA, of river reaches most likely to contain NP. Random samples defined by certain selection criteria were drawn from the EPA River Reach File. Little or no NP was found in river water at most locations (95% < 0.00027 mg/L), while low levels were usually detected in sediment (95% < 0.46 mg/kg). Environmental Risk Criteria and their uncertainties were determined by fitting distribution functions to results of acute and chronic toxicity tests for a variety of organisms. The risk of NP to the aquatic environment is examined by comparing percentiles of observed concentration levels with ERCs using statistical hypothesis tests. Overall risks were calculated by integrating concentration and effect distributions.

077 Detoxification of an Amide-Containing Surfactant by Algae. Tepper, B.E.*, Dunphy, J.C., Pessler, D.G., and Bookland, E.A., The Procter & Gamble Company, Cincinnati, OH; and Hicks, S.L. and Hurshman, B.A., ABC Laboratories, Columbia, MO. Sensitivity of algae to organic chemicals can be influenced by the substance's structure and the algae's response to that substance. Algae have diverse metabolic activity which at times may serve to detoxify chemicals in the environment. In standard toxicity studies with *Selenastrum capricornutum* the surfactant dimethylaminopropyl decanamide (C10 APA) completely disappeared over time. Concurrent with this disappearance was the appearance of polar metabolites. One of these metabolites was identified as dimethylaminopropylamine (DMAPA). Other metabolites were presumptively identified as dimethylaminopropanol (DMAP) and decanoic acid. These observations indicate that algae first cleaved the amide bond in C10 APA resulting in metabolites that are completely biodegradable and at least 100 fold less toxic than the parent substance. This detoxification occurred across a wide range of exposure concentrations, from the NOEC to levels that inhibit algal reproduction by $\geq$ 99%. Rigorous studies demonstrated that the observed metabolism was the result of algae and not the very low levels of bacteria (< 500 cfu/mL) found in test flasks at test termination. Additional studies demonstrated that algae and bacteria metabolize C10 APA by first cleaving the amide bond. These results suggest that algae may play an important role in removing amide-containing organic substances from the aquatic environment. In addition, they suggest that it may be appropriate to incorporate algal transformation into environmental safety/risk assessments of alkyl amides.

078 Environmental Risk Assessment of a Novel Cationic Surfactant Using the European Union System for the Evaluation of Substances (EUSES) Risk Assessment Model. Nordone, A.J.*, Saouter, E., N.V. Procter and Gamble Eurocor S.A., Temselaan 100, B-1853 Strombeek-Bever, Belgium; Tepper, B.E., Federle, T.W., The Procter & Gamble Company, 5299 Spring Grove Avenue, Cincinnati, OH. The environmental risk assessment of a new short chain amide/amine surfactant used in laundry detergent is presented. C8/10 Amidopropyldimethylamine is a water-soluble, non volatile chemical used in laundry liquid detergents. It enhances considerably pre-treat stain removal at low level in product. This chemical is readily and completely biodegrad-able. It passes the "10 day window period" (>60% CO2 production in 10 days) in accordance with the European Directive for the notification of new chemicals. Removal in Continuous Activated Sludge simulation system (OECD 303A - CAS) is above 99%. Removal in wastewater treatment plant is expected to be mainly due to biodegradation. Predicted Environmental Concentrations (aquatic) using the European Union System for the Evaluation of Substances (EUSES) risk assessment model have been calculated using the following three scenarios: a) European tonnage and EUSES default values, b) United Kingdom tonnage, monitored plant emissions and CAS removal rate, and c) United Kingdom tonnage, monitored plant emissions, CAS removal rate, and in stream removal rate. Progression of the scenarios from a to c resulted in a significant reduction in the calculated PEC. These results emphasize the conservative nature of the EUSES default values and the importance of data generation and a tiered approach to risk assessment when using the EUSES model.

079 Characterizing Surfactant Mixture Toxicity Using a Fish Liver Cell Line. Benton, E., Dyer, S.D.*, The Procter & Gamble Co., Cincinnati, OH. A rapid screening procedure has been developed using a fish liver hepatoma cell line (PLHC-1) to determine surfactant toxicity singly and in binary mixtures. Eight surfactants representative of the 3 major classes (anionic, cationic and the nonionic) and dodecanol were evaluated. After determining individual dose response curves (using three cytotoxicity endpoints), 36 different mixture toxicity tests were conducted. Three endpoints were chosen so that modes of action of the chemicals could be evaluated. Neutral Red (NR) uptake was used to determine cell function, Lactate Dehydrogenase (LDH) leakage as an indicator of cytoplasmic membrane function disruptions, and total protein (sulforhodamine blue, SRB) determinations were made as an indication of number of cells/well. Additivity was initially based on concentration addition. In single chemical tests the cationic class was more toxic than both the anionic and the nonionic classes, with the nonionic class (including dodecanol) being slightly more toxic than the anionic surfactants. Predicted additivity curves in general compared well with measured mixture responses. However, when the observed mixture response did not fit this model, independent models and/or isobolograms were then used. In binary surfactant mixtures additivity was generally observed.

080 Biodegradation of Detergent Surfactants: Updating Activities on the Eu Directives. Glod G.*, De Henau H., Procter&Gamble, Strombeek-Bever, Belgium; Masscheleyn, P, Procter&Gamble, ITC, Cincinnati, OH, USA. The process to update the European Union Directive on the biodegradability of detergent surfactants (EU 73/404, 73/405, 82/242, 82/243) has been initiated. Currently surfactants are operationally defined via their response to colorimetric techniques such as the semi-specific MBAS (methylene blue active substances) and BiAS (bismuth active substances) methods. MBAS and BiAS methods are used for anionics and nonionics, respectively and no methods have been implemented for cationics and amphoterics.The currently used OECD confirmatory test does not allow to distinguish between removal and mineralization. The update of the directives aims at addressing essentially the following objectives: (1) improve the methodology for current primary biodegradability directives for anionics and nonionics, (2) expand the scope to amphoteric and cationic surfactants and (3) add an ultimate biodegradability requirement. Based on current discussions between industry (AISE and CESIO), the member states and the EU Commission the following development seems most probable: (a)addition of a surfactant definition to the text and (b) stepwise approach for determining the acceptance of a surfactant in detergents. In a first step a simple screening test on ultimate biodegradability is envisioned. Step 2 assesses primary biodegradability for those surfactants (all classes) that failed step 1 while step 3 assesses in a simple test whether hazardous metabolites may be formed for surfactants that did not pass step 1.

081 Automated Biomonitoring Systems: Opportunities and Challenges. van der Schalie, W.H., U.S. EPA, Washington, DC. Since their inception in the early 1970's, automated biomonitoring systems (biomonitors) have been used to evaluate ambient water conditions, effluent discharges, and drinking water intakes. When linked to automated sampling devices, biomonitors can provide biologically-directed sampling of water or wastewater. Biomonitors provide real-time, continuous monitoring of environmental media using electronic sensors that monitor physiological responses of organisms. These systems can give an early warning of developing toxicity and can detect the presence of unsuspected chemicals or toxic interactions. Recent technological advances offer the possibility of a new generation of biomonitoring systems that can be linked to provide real-time toxicity data over a range of spatial scales (e.g., watersheds or regions). Biomonitors are most effective for detecting sporadic toxic events. However, their sensitivity and response time depend on the organism and endpoint monitored as well as the specific chemicals present. Because responses are sometimes caused by variations in water quality parameters (e.g., temperature, dissolved oxygen, or conductivity), selected physical and chemical parameters should be monitored simultaneously to facilitate interpretation of biomonitor alarms. Other important factors for evaluating the usefulness of biomonitors include the reliability of the system and costs for installation and support. Althougth automated biomonitoring systems have not been widely used for environmental monitoring, their ability to facilitate immediate response and intervention to developing toxic conditions can be invaluable.

082 Experiences with Real World Applications of Automated Biomonitoring Systems. Gruber, D., Biological Monitoring, Inc., Blacksburg, VA. Although all end users desire an automated biomonitoring system which is reliable and sensitive, numerous other factors come into play when trying to apply such systems to the real world. First, all documentation, in addition to the product itself, must reflect a highly competent and experienced commercial operation. Foreign currency, shipping issues, and negotiating letters of credit all must be understood when dealing with foreign countries. The biomonitoring system must require the minimal of installation, training, and operating expenses. The alarm responses must be simplistic (for example, red light vs. green light), they must be readily interpretable, and they must have remote communications compatible with the local installations. All components must be compatible with industrial environments. Sensors (the fish) must be readily available. This presentation will cover numerous issues which make the application of automated biomonitoring systems in the real world a unique experience requiring both patience and expense.

083 Continuous Automated Biomonitoring For Acute Toxicity. Shedd,T.R.*, USACEHR, Ft.Detrick,MD; van der Schalie,W.H., U.S.EPA, Washington,DC; Leach,J.D., USADOIM, Ft.Detrick,MD; Widder,M.W., GEO-CENTERS,INC., Ft.Detrick,MD; Finch,R.A., Gardner,H.S., USACEHR, Ft.Detrick,MD. An Automated Fish Biomonitoring System (AFBS) was developed by the United States Army Center for Environmental Health Research to identify developing toxic conditions in water by continuously monitoring the ventilation and movement patterns of the bluegill (Lepomis macrochirus). This Monitoring System provides an early warning that reduces the risk of causing environmental damage from a release of toxic effluent. The use of continuous biomonitoring was recognized by State and Federal Regulators and quickly embraced as a monitoring strategy. The development of the AFBS included input from researchers, regulators, engineers, programmers, and project site managers for monitoring a complex effluent discharge. Physiological stress to the bluegills, characterized by changes in fish ventilation and movement patterns, is used as an early warning to identify developing acute toxicity of a treated groundwater effluent discharge. An IBM-compatible PC continuously monitors and records ventilatory rate, ventilation depth, cough rate, and whole body movement of up to 32 fish simultaneously. Monitoring begins with 16 fish held in control water for a three-day acclimation period followed by four days of baseline data collection. The fish are then divided into two groups (8 fish-control, 8 fish-effluent). During the subsequent continuous exposure for two weeks to effluent, the computer provides immediate analysis of statistically significant departures from baseline conditions for fish in the control and effluent-exposed groups. The AFBS has been integrated with a Groundwater Treatment Facility. When the monitoring system identifies a potentially toxic effluent, an effluent sample is automatically collected for chemical analysis, a remote monitor in the treatment facility identifies the problem to the facility operators, and if necessary, the discharge is diverted to storage tanks until the problem is resolved.

084 The Use of Physiological and Behavioural Measurements from Invertebrates as Biomonitors of Aquatic Systems. Bamber, S.D.; Depledge, M.H. Plymouth Environmental Research Centre, University of Plymouth, UK. Monitoring various aspects of the physiology and behaviour of selected organisms provides us with the opportunity to gauge the biologically relevant impact of complex effluents entering the aquatic environment. Thus, an integration of all the various potential contributory factors to changes in the status of an individual organism, including physical and temporal as well as chemical factors, can be achieved. The challenge facing us today is to develop biomonitor techniques which detect biologically relevant changes in organisms, in a system which is easy to use, produces an unambiguous reliable signal and requires the minimum of maintenance. At the Plymouth Environmental Research Centre we are currently developing a range of monitoring systems designed to examine variation in higher level biological systems of a diverse range of invertebrate organisms, including crabs, crayfish, daphnids, freshwater and marine mussels and annelid worms. Monitoring the heart rate of various invertebrates, using the non-invasive CAPMON (computer aided physiological monitor) system, has been successfully performed under both laboratory and field conditions and has been shown to provide a clear indication of toxic insult. Behavioural studies have centred around an actograph system which uses infrared light beams to record activities such as tube ventilation in annelid worms, swimming behaviour in daphnids and endogenous locomotor activity in shore crabs. The linkage between simplicity of use of the biomonitor system and the quality and biological relevance of the data it provides appears to be a major factor in the future development of effective biomonitors.

085 Real-time Automated Biomonitoring Using Bivalves. Waller, W.T.*, University of North Texas, Denton, TX, Allen, H.J., University of North Texas, Denton, TX, Ochandio, M.R. , University of North Texas, Denton, TX, Morgan, E.L., Tennessee Tech, Cookeville, TN. Recent developments in real-time automated biomonitoring using bivalves include development of a scheme to characterize bivalve behavior using autoregression and an acute response alarm system. Data from laboratory monitoring of *Corbicula fluminea* was used to characterize baseline behavior and an acute response. Data from field monitoring were used to further define baseline behavior and the acute response alarm. Technology related issues faced in the deployment of remote biomonitoring platforms are discussed including data analysis, and telemetry.

086 The New "Multispecies Freshwater Biomonitor" for Ecological Relevant Control of Water Quality. Gerhardt, Almut; LimCo International, Ibbenbüren, Germany. A new automated biomonitoring system is developed based on quadropole impedance conversion technique. This universal measuring principle allows to register simultaneously different types of behaviour of all kinds of freshwater organisms. The measuring unit contains two pairs of steel electrode plates, one producing an alternating current, the other measuring the impedance changes due to movements of the organism in the chamber. Each electrode pair is attached to the respective opposite walls of the test chamber, which is made of cylindrical or rectangular plexiglas with the test water flowing through. According to the special ecological demands of the organism, sediment and detritus is added, without affecting the signal quality. Signals from up to 96 test chambers can be measured and analysed online. The following parameters are registered: survival, ratio of activity/inactivity, different behaviours as defined by different frequency and amplitude patterns, such as locomotion and ventilation. Time series analysis for each parameter generate the model behaviour, deviations from which result in a behaviour-specific alarm. The alarms for different behaviours as well as for the different test species can be graded according to different sensitivities and response thresholds. This leads to an ecological relevant and reliable 'water quality alarm'. Early warning responses have been measured within 60 min. in different invertebrates exposed to different kinds of water pollution: Gammarus pulex (Crustacea) reacted with increased ventilation and decreased locomotion within 60 min. to a copper effluent of 3-100 mg/l. Hydropsyche angustipennis (Insecta) reacted to a complex industrial effluent within 90 min. with decreased ventilation and increased locomotion. Changes in behaviour due to toxic water have also been found for Daphnia sp., chironomids, plecopterans and fish. The "Multipsecies Freshwater Biomonitor" is a sensitive, ecological relevant biological early warning system with universal applications.

087 Gymnotox, a New Warning Biomonitor Using a Neotropical Electric Fish, *Apteronotus albifrons*: Principle and Performance. Thomas, M., International Center for Water, Nancy, France. Water is constantly subject to ever increasing risks of pollution and more stringent quality requirements are imposed on water destined for human consumption. Therefore water resource managers have set up early warning stations which are equipped with physico-chemical sensors and analyzers to monitor specific parameters. Biological monitoring techniques are also added because they have the advantage of being non-specific and consequently provided overall information on the quality changes of the environment. It is within this framework that a new biomonitor, named Gymnotox, has been developed by the NAN.C.I.E. and the Laboratory of Applied Biology (Nancy1 University). Gymnotox exploits the Electric Organ Discharge (or EOD) emitted by a neotropical fish, *Apteronotus albifrons* (Gymnotiformes) and uses the fact the EOD frequency varies as a function of water quality. The first objective is to describe this new biomonitor especially the biological reagent, the hydraulic system, the data recording, processing and analyzing with the supervisor Ramscope. The data are analyzed in real-time, where each fish represents its own standard. The second objective is to present the influence of non-toxic physico-chemical parameters (temperature, pH and conductivity) and pollutants (cyanide and phenol) on the electric behavior of *A. albifrons*. The results show that EOD frequency is temperature (45.4 to 63.9 Hz/°C) and pH dependent whereas stay stable with different ionic charges (54 to 484 $mC.cm^{-1}$). Concerning the pollutants, the KCN and phenol detection thresholds were found to be 35 $mg.l^{-1}$ and 1 $mg.l^{-1}$ respectively. An important advantage of Gymnotox resides in the use of the remarkably stable electric information generated directly by the fish, which can be likened to an electric generator. The electrical behavior recording and processing are therefore easier. The performance to detect water quality changes is also particularly interesting. For these reasons, Gymnotox seems to be an appropriate mean to monitor water quality.

088 Sensor Technologies for Future Aquatic Automated Biomonitoring. Sarabun, C.C.*, The Johns Hopkins University Applied Physics Laboratory, Laurel, MD; Ondercin, D.G., The Johns Hopkins University Applied Physics Laboratory, Laurel, MD. The Johns Hopkins University Applied Physics Laboratory (JHU/APL) is developing technologies under a number of DOD programs which could be applied in future biomonitoring programs. These technologies include sensors, and signal and information processing techniques. Sensors under development at JHU/APL include those which can monitor bioindicators directly or whose adjunct data can significantly affect the interpretation of primary biomonitor data. Sensors under development include biologically directed MicroElectroMechanical (MEM) sensors which have been used to detect nerve agents, polymer matrix-based heavy metal specific sensors which have been configured to detect lead and uranium, miniaturized time of Flight spectrometers for organic materials, low-noise, high-sensitivity electric and magnetic field sensors and specialized, fast response fiber optic sensors used, for example, to detect nitrates and phosphates. Signal and information processing techniques include techniques for feature and signal extraction, signal analysis of multiple parameter data streams and AI-assisted decision making aides. Some of these approaches are currently being applied to the analysis of fish ventilatory data. This paper will discuss some of these flexible technologies and the ways in which they have been applied and ways they might be applied in the future to enhance biomonitoring capabilities.

089 Remote Applications of Automated Biomonitoring to Watershed Drainages. E. L. Morgan*, E. T. Ososanya, D. B. George, Tennessee Technological University, Cookeville, TN, and W. T. Waller and H. J. Allen, Institute of Applied Sciences, University of North Texas, Denton, TX. Three decades of progress have been made since John Cairns and his associates first coined a new scientific endeavor known as automated biomonitoring. Development and application have ranged from initial systems designed for early warning of wastewater discharges using fish as sensors, to configurations for automated bioassays using bivalve mollusc or other invertebrates. More recently, remote biosensing devices have been implemented in watershed networks instrumented with water quality monitoring stations and satellite data retrieval. Presently, a new generation of intelligent biosensing systems are being developed for near, real-time continuous monitoring at remote river platforms. One system built in collaboration with the University of North Texas and U.S. EPA uses digital signal processing and fast fourier transforms to generate power spectrum densities (PSD) that represent aquatic animal functions, i.e. cardiac and breathing patterns. PSD are used as functional features for input into artificial neural networks (ANN). Once ANN are trained using stressed and non-stressed patterns, the system can judge adverse water quality and provide a control sequence capable of triggering an automated water sampler. The control sequence can also alert concerned parties so that samples can be retrieved and processed for causative agents, and corrective action taken when warranted.

090 Automated Biomonitoring Systems: Current Status and Future Development. Dickson, K.L., Waller, W.T., Morgan, E.L. This presentation will be the last of the talks in the Biosensor Session and will summarize the current status of biosensing, including the information from the talks in the session. Additionally, the author will conjecture as to the future direction and role of automated biomonitoring systems in water quality monitoring and protection.

091 The Complex Link Between Public Policies, Human-Induced Land Use Change, Ecological Risk and Social Benefits. Bockstael, N.*, U. Maryland; Leggett, C., U. Maryland, College Park, MD. We explore the two-way link between ecological risk and natural resource economics by first tracing the human decisions about land use that alter physical and biological stressors in the adjacent marine environment and then by evaluating some of the consequences to humans of the resulting ecological change. The principal focus of the research is how public policies can be formulated in such a way as to affect human decisions about land use. Ecological models can trace the resulting land use pattern to water quality consequences. We show some of the ways in which these water quality consequences feedback to effects on human welfare. The focus, then, is on how policies that alter human decisions about land use can have ecological risk consequences that ultimately affect society's welfare.

092 Use of the "Service-to-service" Approach to Address Ecological and Recreational Service Losses For a Freshwater Oil Spill Natural Resource Damage Assessment (NRDA) in Reston, Virginia: Case Study. Nicolette, J.P.*, ENTRIX, Inc. and Nicolette Environmental, Inc., Grayslake, IL; Markarian, R.K., ENTRIX, Inc., Wilmington, DE; Pfeifer, C.E., ENTRIX, Inc., Wilmington, DE; Rockel, M.L., ENTRIX, Inc., Wilmington, DE; Pearson, D.V., Colonial Pipeline Company, Atlanta, GA. On March 28, 1993, a subsurface pipeline ruptured and discharged approximately 400,000 gallons of No. 2 fuel oil (diesel) into the environment. The discharged oil flowed overland to nearby storm sewers that empty into Sugarland Run, a freshwater tributary to the Potomac River. As the oil flowed downstream through aquatic and riparian habitats, it injured ecological and recreational natural resources and services associated with those habitats. The natural resource damage assessment process was utilized in assessing ecological and recreational injuries resulting from the spill and in developing appropriately scaled restoration of the injured resources. Ecological service losses (injury) were scaled to restoration through the use of the habitat equivalency analysis (HEA) model. Recreational service losses were scaled to restoration utilizing economics methods (e.g., modified benefits transfer). In this case and in meeting the goal of the OPA NRDA Rule, the responsible party agreed to compensate for the lost ecological and recreational services resulting from the spill, by providing through a restoration program, ecological and recreational services (*service-to-service* approach).

093 Expedited Resolution of NRD Claims for Injury to Freshwater Wetlands- The Bennington Model. Hazardous waste releases from the Bennington Landfill Superfund Site, including PCBs and heavy metals were identified by the natural resource trustees (DOI and Vermont Agency for Natural Resources) as resulting in injuries to forested wetlands and ponds. A habitat equivalency analysis based on site specific sampling data tied to toxicity reference values was employed to evaluate past, interim and future losses using an acre-year metric, and identify restoration requirements. This methodology also permitted the trustees to factor into claim development the fact that remediation by EPA was anticipated at only some of the areas of concern to the trustees. The PRPs were then invited to participate in identifying candidate restoration projects. Working with the Town of Bennington, a site PRP, an innovative restoration package was developed which called for reestablishment of natural hydrologic conditions and wetland plant communities at an old cistern-fed water supply complex, protection of surrounding habitat, construction of facilities for public access, and development of an educational program tied to the local school system, which was then implemented by the Town. This approach permitted expedited resolution of trustee NRD claims while minimizing transaction costs to all parties. National application of this approach, which is potentially applicable to numerous hazardous waste sites, would permit natural resource trustees to achieve effective restoration more rapidly and at lower cost than through traditional assessment techniques.

094 Developing a Framework for the Economic Valuation of Ecological Benefits. Blake-Hedges, L.*, U.S. EPA, Washington, DC; Harris, J., U.S. EPA, Washington, DC; Kibler, V., U.S. EPA, Washington, DC. The EPA Social Sciences Discussion Group, which is convened under the auspices of the EPA Science Policy Council, is working to improve the Agency's ability to conduct comprehensive cost-benefit analyses. This presentation describes one part of this effort, the development a conceptual framework that sets out commonly used methods for conducting ecological benefit assessments. The purpose of this Framework is to provide ecological risk and economic benefit valuation information to enhance the Agency's ability to incorporate ecological benefit considerations in its risk management and pollution prevention policies. It identifies tools and information resources for ecologists and economists involved in ecological benefits assessment. The framework, intended for a wide audience, provides background on the conduct of ecological benefit assessments, including discussions on ecological risk assessment and ecological benefit valuation techniques. It discusses some of the difficulties in developing ecological benefits assessments and identifies the need for coordination and interaction between scientists and economists. The Agency anticipates developing a workshop to discuss the framework and to identify research needs, case studies, and specific areas where interdisciplinary coordination can be most beneficial in the development of ecological benefit assessments.

095 Spatial Weight-of-Evidence Integrated Risk Assessment. Rogers, W.J.*, West Texas A&M University, Canyon, Tx; J.W. Bickham, Texas A&M University, College Station, Tx; T.M. BolwahOnn, Battelle Memorial Institute, Amarillo, Texas. The *spatial weight-of-evidence integrated risk assessment* approach uses traditional risk assessment methods but adds a holistic approach by translating ecological effects into gain or loss of habitat value, or *weighted usable habitat.* Habitat value is based on an easily understood scale from 0.0 to 1.0 (1 being ideal habitat). The habitat value is supported by numerous measurable variables that are translated into one habitat or resource value. The approach integrates traditional contaminant fate and transport modeling typically used in remediation investigations with resource valuation methodologies and models typically used in natural resource assessment. This spatial approach provides (1) a measure of weighted usable habitat under the current and future no action alternative and (2) a method to determine net ecological benefit and impact of the potential remediation alternatives. Potential for replacement of lost habitat value can also be considered and is based on mitigation by acquisition or development of like habitat value. Most natural resource trustees accept the use of *habitat equivalency* methodologies but may lack the resources to complete such an evaluation. The agencies may then elect a simpler, more arbitrary process such as *contingent value*, which can result in excessive natural resource claims. The integrated approach expands the scope of the traditional ecological risk assessment by providing a tool to support the decision-making process to address multiple stakeholder concerns.

096 Valuing Ecological Services Using an Ecological Metric to Facilitate the Evaluation of Site Closure and Corrective Action Options. Rockel, M.L.*, Kealy, M.J., Tomassi, T., Friant, S.L., and Markarian, R. ENTRIX, Inc., Wilmington, De. Cost/benefit analysis, the traditional framework for organizing information on the economic effects of an action does not indicate whether or not the environment is improved or diminished by an action. However, the net effect on the environment is an issue when managing ecological risks. Net Environmental Benefits Analysis is proposed as an organizing framework for summing the positive and negative ecological effects of one or more proposed risk reduction remedial alternatives. NEBA uses an ecological metric, or currency and an economic model that quantifies the amount of currency that is lost or gained over time, enabling the ranking of risk management activities with respect to their net benefit to the environment. In the context of Superfund and RCRA legislation, historically preferred remediation activities include: dredging contaminated sediments from a stream or wetland; capping contaminated soils; pumping groundwater to remove or contain contaminants; and other engineering-based solutions to pollution. While such remediation activities can effectively prevent contaminants from causing harm to human health or the environment, in some cases, they can also harm the environment by changing an essential aspect of a habitat. The present paper demonstrates how to integrate NEBA into the ecological risk assessment/management process in the Superfund and RCRA contexts to assist in the selection of remedial alternatives. Results from case studies illustrate the utility of the approach.

098 Ecorisk and Nrda Endpoints: Some Are, Some Aren't. Williams, B.A.,ENTRIX, Walnut Creek, CA; D. Haury, ENTRIX, Chicago, IL; J.A. Holder, ENTRIX, Walnut Creek, CA, and G.A. Robilliard, ENTRIX, Seattle, WA. The Natural Resource Damage Assessment (NRDA) process is used to determine the damages owed to a public trustee for natural resource injuries that occurred as a consequence of an action, usually an unauthorized released hazardous substance or oil. The stressor and its adverse effects are usually known in an NRDA. The primary driver of the NRDA process is determining if quantifying the loss of services provided by the natural resource and in many cases determining the economic value of those lost services. In contrast, an Ecological Risk Assessment (EcoRA) is used to assess the probability of future harm to the environment as a consequence of a specific action. Both processes require identification of 1) stressors; 2) affected biological receptors; and 3) exposure scenarios. Although the basic ecological considerations of each process are similar, significant differences in economic and temporal focus could result in divergent, or even conflicting products. However, up front planning between Natural Resource trustees and EcoRA managers can identify the overlap between the two processes. This can result in a more cost efficient and scientifically defensible damage assessment. For example, while the EcoRA process has no provision to assess the economic impact of predicted adverse effects, the quantification of adverse effects in an EcoRA could feed into the estimates of lost services and thus damages in the NRDA. A case study of the simultaneous implementation of an NRDA and EcoRA for a closed refinery site in Texas provides a means to compare and contrast the similarities and differences between the two processes. The ecological, economic, measurement, and management goals and techniques used to simultaneously conduct both an NRDA and an EcoRA are presented.

099 Integrating Risk Assessment and Damage Assessment: Is There a Common Ground?
Chapman, D.J.,* Gouguet, R.G., Kern, J.C. NOAA. This paper focuses on the relatively novel approach in Natural Resource Damage Assessment of coordinating risk assessment and damage assessment studies for CERCLA site investigations. Quantitative Risk assessment methods used in the CERCLA RI/FS process, focuses on developing measures of potential harm to humans and ecological receptors with respect to contamination concentrations in order to determine clean up levels. Natural Resource Damage Assessments focus on developing the appropriate type and scale of restoration for quantified injury to resources and/or services. Risk assessment studies can provide valuable information for use in NRDA. Potential time and cost benefits exist if studies designed for risk assessment and damage assessment can be coordinated and/or combined with the NRDA process. Using case examples, this paper outlines how risk assessment investigations can be used, or modified, to identify and quantify the necessary NRDA endpoints of scaling restoration.

100 HERMES: Methods for Including Ecological Values in Environmental Restoration. Scott, M.J.*, Pacific Northwest National Laboratory, Richland, WA; Bilyard, G.R., Pacific Northwest National Laboratory, Richland, WA; Sackschewsky, M.R., Pacific Northwest National Laboratory, Richland, WA; Tzemos, S. Pacific Northwest National Laboratory, Richland, WA; Walker, B.A., Washington State University, Pullman, WA. Pacific Northwest National Laboratory scientists have developed a Geographic Information System (GIS) based computer decision support tool known as Health and Ecological Risk Management and Evaluation System (HERMES). This tool provides a concise description of the current environmental landscape that can be used to evaluate the ecological and monetary tradeoffs between future land use, restoration and remediation options before action is taken. Ecological impacts that are evaluated include effects to individual species of concern, habitat loss, and habitat fragmentation. Monetary impacts include those associated with habitat mitigation. More complex economic and ecological interactions can be incorporated into HERMES. In developing HERMES, we captured economic values associated with the ecological resources in a shrub-steppe dryland habitat being displaced by development or cleanup actions. Several resource valuation tools from environmental economics were used, including benefits transfer. The highest values of natural shrub-steppe habitat appear to be derived from soil stabilization. Key data include types of ground cover, types of habitat, and locations of species of concern (threatened, endangered, or candidates for listing). The HERMES code can be used to evaluate the extent and types of habitat and species disturbance associated with hazardous waste mitigation projects, and to help determine actions that must be taken to reduce ecological risks of mitigation, together with their costs.

101 Silver (And Other Metals) at the Regulatory Crossroads: Good Scientific Focus Is Needed to Answer Pertinent Questions. Purcell, T.W.*, The Silver Council, Chevy Chase, MD. Silver enters the environment from a variety of industrial, domestic and natural sources. Because of these inputs, there exists the possibility for exposure of ionic and complexed silver to terrestrial and aquatic organisms, as well as to humans. This potential for exposure has resulted in promulgation of a number of regulations in the United States, beginning in the 1960s. Rules were first developed to protect against possible negative effects to humans due to ingestion of drinking water treated with silver. As regulations for other environmental media were developed, silver was generally included because of its designation as a chemical of concern in drinking water. Ambient Water Quality Criteria were calculated from laboratory data based on the toxic action of ionic silver. In the early 1990s, the U.S. Environmental Protection Agency Office of Drinking Water downgraded silver from a primary to a secondary maximum contaminant level because the effects of silver exposure on humans were judged to be cosmetic rather than toxic. Regulations pertaining to solid waste, surface waters and occupational exposure have not changed based on recent scientific work, but may do so in the near future as the fate and toxic mechanisms of this metal, along with others, become better understood. Silver is also expected to be included in upcoming sediment quality criteria guidance. Scientific information is needed to fulfill recommendations provided by the SETAC Pellston Workshop in *Reassessment of Metals Criteria for Aquatic Life Protection* (Bergman and Dorward-King, SETAC Press, 1996). Data addressing environmental interactions and toxic mechanisms is required to answer questions about the bioavailability of silver and other metals so that future regulations can be crafted accurately.

102 Detection of Ag+ Ion Utilizing a Silicon-Based Potentiometric Sensor. Chyan, O.*; Chen, J.J.; Xu, F.; Gao, J.; University of North Texas, Denton, Texas 76203. Recently, a great deal of research effort has been dedicated to determine the sources, transport, and fate of silver in various aquatic environments. Several researchers have investigated silver toxicity in freshwater systems and concluded that the free silver ion, Ag+, has the highest toxicity among different chemical forms of silver . Therefore, it is highly desirable to measure trace levels of free silver ions in natural water systems. In this paper, we will discuss a new silicon-based sensor assembly which was shown to sensitively detect free silver ions (at below parts-per-billion level) in several water samples. The Ag+ detection capability is accomplished by a direct measurement of the open-circuit potential from the silicon-based sensing electrode using a high input impedance potentiometer. Our surface spectroscopic data and atomic force microscopic images indicate that the silicon space-charge region influenced by the Ag+ containing solution contributes to this unique Ag+ sensing capability. In addition to the sensor's operation mechanism, the sensor's performance and its limitations will be discussed.

103 Silver Thiolate Chemistry: an Overview of the Solid State and Aqueous Solution. Bell, R.A., Bennett, S.E., Britten, J.F. and Kramer, J.R. Department of Chemistry, McMaster University, Hamilton, ON, Canada. The structure and chemistry of silver(I) complexes of environmental mercaptans is central to our understanding their mobility and fate. Regulation should be based on this understanding. Environmental silver thiolates act as transient carriers of silver(I) because of their aqueous solubility and their ability to transfer silver(I) to organic and inorganic colloidal matter containing sulfur groups. We propose that silver(I) thiolates themselves exist as colloidal aggregates composed of short, interlocking chains of -S(R)-Ag-S(R)-Ag- units which are continually breaking and reforming. We suggest that silver is very mobile among different mercaptans, and the presence of any HS^B will result in the eventual formation of Ag_2S. We have used x-ray powder diffraction (XRD) and single crystal x-ray analysis together with nuclear magnetic resonance (NMR) and electrospray mass spectrometry (ESMS) to study the environmental silver thiolates, cysteinato-silver and 3-mercaptopropanoato-silver. Our synthesis of cysteinato-silver has resulted in a microcrystalline solid, the XRD pattern of which is consistent with a layered structure. 3-Mercaptopropanoato-silver, crystallized for the first time, has a layered structure, which is developed from in-plane one dimensional chains of -S(R)-Ag-S(R)-Ag- zig-zags as the fundamental units. ESMS showed the presence of a range of multisilver-multiligand species. Proton and carbon-13 NMR of aqueous penicillaminato-silver, a planar structure made up of *double-helix* chains, shows broad lines at room temperature which is ascribed to aggregate formation (n = 20 - 60 in $[RS\text{-}Ag]_n$). A variable concentration study shows rapid ligand exchange. Addition of HS^B to silver(I) thiolate solutions quickly converts these compounds to Ag_2 S.

104 Fate of Silver in the Environment: Hypotheses. Kramer, J.R.*, Bell, R.A., McMaster University, Hamilton, ON.. Silver forms simple inorganic sulfides ($AgHS^0$), thiolate and polysulfides in natural waters. Silver, present at low ng L^{-1} levels in most instances, forms simple inorganic and thiolate complexes. Furthermore, most of the aqueous silver is in the colloidal (<0.45mm) fraction. This colloidal material is predicted to persist as multiple chain (layered) relatively low molecular weight substances, probably sorbed to natural organic matter. Furthermore this fraction persists metastably in oxidized water. Bivalves accumulate silver. They are predicted to take up colloidal Ag-thiolates and form stable Ag-thiolates or Ag-sulfide in the exterior cells. The cells are later sloughed off, and the Ag(I) in the residue is forms a more inert Ag-sulfide. Key unknowns are the specific nature of Ag(I) in the colloidal fraction, size distributions as related to environmental factors and the bio-assimilation of Ag-thiolates, the role of other metals (e.g. Fe(II) and Fe(III)) in development of macro-molecules containing Ag(I), and the protective effect against Ag(I) migration by thiols in the bivalves. A scheme for measurement and interpretation of these parameters is proposed in context to modeling and regulatory use.

105 Silver in Colorado Watersheds - Concentrations and Chemical and Phase Speciation. Gill, G. A., Wen, L.-S.*, Santschi, P. H., Tang, D., and Lehman, R., Texas A & M University at Galveston, Galveston, TX. Surface river water samples and municipal and industrial discharge effluents were collected using ultra-clean sampling protocols at 5 sites in the State of Colorado. Sampling was conducted above and below an effluent discharge to characterize silver discharges on the receiving water body. All samples were analyzed for total (unfiltered collections), filtered (0.1 and 0.4 mm), particulate (particles on 0.45 mm filters) silver, and two colloidal silver size fractions (3 kDa B 0.1 mm and < 3 kDa) using ultra-clean sample preconcentration protocols. In general, upstream unfiltered and particulate silver concentrations fell in a fairly narrow range, 3.1 to 21 ng/L and 0.2 to 1.7 mg/g, respectively. Downstream unfiltered and particulate silver concentrations were more broad, 2.8 to 1110 ng/L and 0.5 to 104 mg/g, respectively, and reflected impacts of silver laden discharge effluents. The filtered samples general mirrored that observed for the unfiltered and particulate samples, but at lower overall concentration levels. The dominance of particulate matter and chloride on the phase and inorganic solution speciation of silver was pronounced. Using thermodynamic equilibrium modeling, free silver ion concentrations varied from a high of 72% of the total silver present, when both TSS and chloride ion content were low, to a minimum of 0.4%, where chloride and TSS were both abundant. Cross-flow ultra-filtration revealed that, on average, more than 60% of the filtered silver (0.1 mm) was associated with colloidal macromolecular organic matter; 16 to 63% of the 0.1 mm filtered silver concentration was in the size range between 3 kDa and 0.1mm, and 9 to 44% of the filtered silver (0.1 mm) was < 3 kDa.

106 The Reaction of Silver and Sediment AVS: Ag_2S Formation, Measurement, SEM Determination and Possible Oxidation Pathways. Mahony, J.D., Di Toro, D.M., McLeod, P., Morrissey, J., Santos, L., Manhattan College, Riverdale, NY. The formation of Ag_2S in the pure phase as well as in sediment follows the same general pattern of other heavy metal sulfides. The kinetics of this process will be presented and compared with that of other heavy metal sulfides. The measurement of AVS when Ag_2S is present offers some unique problems as does the determination of SEM. Various approaches to resolving these difficulties will be presented. The correlative property to the formation of all heavy metal sulfides is their oxidation rates. Because of the importance of this with respect to fate and transport modeling, several different approaches have been taken to investigate this process. These include varying the oxidant by varying certain components commonly found in sediments. The results of these studies as well as their application to a sediment flux model will be presented.

107 Silver Toxicity to *Chironomus tentans* in Two Freshwater Sediments. Call, D.J.*, Polkinghorne, C.N., Markee, T.P., Brooke, L.T., Geiger, D.L., Lake Superior Research Institute, University of Wisconsin-Superior, Superior, WI., U.S.A. Sediment from two freshwater lakes was amended with $AgNO_3$ to determine (1) amendment levels needed to exceed the Ag binding capacities of the sediment for the occurrence of measurable concentrations of *dissolved Ag* in pore water (PW), and (2) the toxicity of amended sediments. *Dissolved* Ag was procedurely defined as the Ag that passed through a 0.2 Fm membrane filter. Ten-day toxicity tests were performed with *C. tentans* larvae first without sediment present to obtain a *water-only* LC50 value, followed by $AgNO_3$-amended sediment exposures designed to yield PW dissolved Ag concentrations that bracketed the water-only LC50. Both sediments contained levels of AVS and TOC less than 1.2 Fmol/g and 1.4 percent, respectively. West Bearskin Lake sediment, which contained a larger proportion of fine particles (silt and clay) and had a higher iron content than Bond Lake sediment, was more effective in binding Ag, as amendment levels in excess of 2.98 and 0.080 g Ag/kg dry sediment, respectively, were required before appreciable concentrations of dissolved Ag appeared in the PW. These concentrations of Ag are considerably higher than typical reported field concentrations of <0.005 g/kg. The 10-day LC50 value for dissolved Ag of 0.057 mg/L in water without sediment present was compared to LC50 values obtained following Ag incorporation into the two sediments. The higher Ag amendment levels resulted in the displacement of Zn, Ni, Pb and Cu from sediment to PW, with Zn attaining the highest concentrations. Larval mortalities in these exposures were likely due to a mixture of metals. The capacity of these sediments to bind Ag appears to have exceeded the combined binding capacities of their AVS and TOC levels, indicating that additional binding phases may also have been operative.

108 Fate and Effect of Silver Compounds on the Anaerobic Digestion Process. Pavlostathis, S.G.*; Maeng, S.K., Georgia Institute of Technology, Atlanta, GA. Laboratory experiments were conducted to assess the anaerobic biodegradability of a silver-bearing, waste activated sludge as well as the effect of silver compounds on the anaerobic digestion process. All assays were performed at 35°C in the dark. The ultimate biodegradability of silver-bearing waste activated sludge (5.0 g silver/kg sludge solids) was 61% as compared to 59% for the control (i.e., silver-free) sludge. The rate and extent of methane production was similar for both sludge samples. Addition of either silver nitrate or silver sulfide to methanogenic, mixed cultures up to an equivalent concentration of 100 mg Ag/L did not affect the rate and extent of methane production. Silver thiosulfate when tested at an equivalent concentration of 100 mg Ag/L (and 1000 mg S/L), resulted in accumulation of ca. 28 mM of fatty acids (mainly acetate), 90% inhibition of methanogenesis and 39% inhibition of acidogenesis. However, using silver-free, thiosulfate-amended controls, it was concluded that the observed inhibition was not contributed to the silver but rather to the excess thiosulfate (used as an alternate electron acceptor resulting in the production of soluble sulfide at inhibitory levels). Computer simulations using MINTEQA2 resulted in extremely low concentrations (< 10^{-15} M) of free silver ions (Ag^+) which explains the lack of inhibitory effects due to silver. The two predominant silver species were Ag_2S and Ag^o. In conclusion, due to the high complexing capacity of the anaerobic digester mixed liquor as well as the reduction to elemental silver, relatively high concentrations of silver (at least up to 100 mg Ag/L) can be tolerated by anaerobic digestion systems. The results of this study have implications on the biological treatment and management of photoprocessing wastewaters.

109 Interactions of Silver with a Unicellular Alga: Transport Mechanisms Through the Cell Wall and the Plasma Membrane in Relation to Chemical Speciation. Fortin, C.* and Campbell, P.G.C., INRS-Eau, Université du Québec, Sainte-Foy, QC, Canada. Many of the apparent exceptions to the Free-Ion Model (FIM) of metal toxicity involve either ligands that are assimilable on their own (so-called "accidental" metal transport - e.g., *Water Res.* 32: 419-429 (1998)), or ligands that form neutral lipophilic metal complexes (e.g., *Environ.Sci.Technol.* 28: 1781-90 (1994)). In the present project, we are attempting (i) to determine whether enhanced silver uptake may occur in the presence of chloride and (ii) to elucidate the mechanism(s) responsible for this behavior. We have chosen a unicellular alga, *Chlamydomonas reinhardtii*, as our test organism. This species is available in strains with / without the normal algal cell wall, allowing us to quantify the role of the cell wall in silver-algae interactions. Short-term (#30 min) silver uptake is determined using ^{110m}Ag as a radio-tracer in defined inorganic media, with differentiation between adsorbed and intracellular metal. After first quantifying silver uptake rates in chloride-free media, we then increased the Ag and Cl concentrations together, in proportions calculated to give a constant free Ag^+ concentration. In such an experiment, the Free-Ion Model predicts that metal uptake should be constant, i.e. that the biological response should be insensitive to the increase in $AgCl_n$ species in solution. Contrary to this prediction, as [Cl^-] was increased from 0 to 4 mM, at constant ionic strength and constant free [Ag^+] (10 nM), the Ag uptake rate more than doubled (2.4-2.5x). We suspect that this exception to the FIM is caused by passive diffusion of the neutral species $AgCl^0$ and experiments to confirm this interpretation are currently underway.

110 A Probabilistic Tropic Level Model for Silver Bioaccumulation in Aquatic Systems. Warila, J., Batterman, S.*, University of Michigan, Ann Arbor, MI. A probabilistic three tropic level simulation model was developed and tested for the purpose of simulating the bioaccumulation of silver in fresh water aquatic species. Trophic levels include freshwater algae, insects/crustaceans, and trout/carp. Each tropic level considers uptake from water, food and sediment, with first-order uptake and elimination kinetics, typical feeding rates, organism sizes, etc. Most of the parameters in the model, based on a literature review and statistical hypothesis testing, are considered to be lognormally distributed; some parameters follow other distributions. A Monte Carlo simulation reflects parameter variability on the model output, silver dose in each species, in several scenarios representing silver discharges in a range of aquatic ecosystems, including the use of silver as an element of a secondary disinfectant formulation that is discharged in a warm receiving body. The simulation model reproduces results obtained for individual organisms and experimental food-chains with approximate bioconcentration factors of 10^3 to 10^6 for algae, 10^3 for crustaceans and insects, and 10^1 to 10^2 for freshwater fish. The model also predicts results obtained in natural systems, although many model inputs are poorly estimated. Critical variables are identified using sensitivity analyses for each scenario. Under most of the tested scenarios, bioaccumulation of silver appears unlikely to result in toxic effects to aquatic wildlife and humans consuming fish. However, bounding scenarios can result in chronic impacts in some species, e.g., rainbow trout. The model provides a means to reflect the variability in data sets and to estimate the likelihood of various concentrations. This represents a significant advance over simpler rate coefficient models, and results of most likely or 50th percentile predictions from the probabilistic model sometimes differ significantly from that of the deterministic models.

111 Organochlorine Compounds and Hydrocarbons Effect on the Benthic Fauna of Chetumal Bay, Mexico. Salazar-Silva, P.; Noreña-Barroso, E.; Zapata-Perez, O. and Gold-Bouchot, G*. CINVESTAV Unidad Merida, Merida, Yucatan, Mexico. To evaluate the environmental health of Chetumal Bay, a bay located on the border between Mexico and Belize and breeding ground of the West Indies manatee (*Trichechus manatus*), sediment samples were collected on eight stations. Five replicates were collected for benthic macrofauna and one for chemical analysis for each station. The samples were sieved (1 mm) and preserved. The organisms were identified to family, counted and weighed. The usual community parameters were calculated: diversity (Shannon-Wiener and Simpson indices), evenness, number of families and individuals. Significant correlations ($P < 0.05$) were found between EDDTs and number of families ($r = -0.81$), number of families ($r = -0.75$) and the Shannon-Wiener index ($r = -0.81$); between Mirex and evenness ($r = 0.70$); Endosulfan II and number of individuals ($r=-0.72$) and total pesticides and number of families ($r = -0.75$). Using the Abundance-Biomass Comparison (ABC) curves, three stations can be classified as moderately polluted. The BIOENV procedure found a significant correlation ($r = 0.75$) between the distance semi-matrix calculated with the faunal data and the distance semi-matrix calculated with contaminant data with EDDTs, Mirex, EPCBs and salinity. From the analysis it can be concluded that there is a severe impact of organochlorine pesticides on the benthic fauna in Chetumal bay.

112 Predicting Benthic Impacts from Measures of Bulk Sediment Contamination in Estuaries of the Southeastern United States. Hyland, J.L.*, NOAA, Charleston, SC; Van Dolah, R.F. and Snoots, T.R., SC Department of Natural Resources, Charleston, SC. Sediment contaminant concentrations and macroinfaunal community structure were measured synoptically at 242 estuarine sites during the summers of 1994-1996 as part of the Environmental Monitoring and Assessment Program (EMAP) in the Carolinian Province (Cape Henry, VA to St. Lucie Inlet, FL). Relationships between the incidence of a degraded benthos (low number of species, H', abundance, or benthic index score) and several measures of overall sediment contamination were determined in efforts to develop an empirical framework for evaluating risks of benthic impacts from exposure to contaminant mixtures in these estuaries. One measure of contamination was the mean Effects Range-Median (ER-M) quotient: i.e., the mean of the ratios of individual contaminant concentrations in a sample relative to respective ER-M values. Our results showed that mean ER-M quotients at sites with a degraded benthos ranged from 0.005 to 0.438 and that 50% of these samples had quotients of 0.052 or less (based on a best-fit curve applied to the data). No sample with a degraded benthos had a corresponding ER-M quotient > 1.0, the beginning of the range for Òhighly toxic samplesÓ based on lab toxicity studies and a broader national database (Long et al. 1998). Thus, for southeastern estuaries, ER-M quotients ranging from about 0.05 to 0.5 appear to be indicative of ÒhighÓ sediment contamination associated with a relatively high incidence of benthic impacts. Most samples with healthy benthic assemblages (92%) had mean ER-M quotients < 0.05. These results provide a useful framework for predicting benthic impacts in future assessments based on empirically derived animal-contaminant relationships from field samples.

113 Evaluating the Bioavailability of Metals Mixtures in Sediments from the Clark Fork River Basin. Naddy, R.B.*, Stubblefield, W.A., Christensen, K.P., Pillard, D.A., Tucker, S.A. and Hockett, J.R. ENSR, Fort Collins, CO. Due to historic mining activities in southwestern Montana, several metals, e.g., As, Cd, Cu, Pb, and Zn, have been released into adjacent aquatic habitats (i.e., Silver Bow Creek and the Upper Clark Fork River). Presently, remedial activities include liming of waters from Silver Bow Creek and retention in settling ponds (Warm Springs Ponds), to reduce metals concentrations prior to discharging into the headwaters of the Upper Clark Fork River (UCFR). Several studies have been conducted to-date to evaluate the potential bioavailability of these metal mixtures from sediments in both the Warm Springs Ponds (WSP) and the UCFR. Bulk sediment and sediment porewater metals concentrations were measured and compared using appropriate Sediment Quality Criteria methods (i.e., SEM-AVS and sediment porewater summed as toxic units [i.e., IWCTU]). Sediments in the UCFR were sampled in both depositional and riffle habitats. Porewaters were sampled through the use of *in situ* peepers, buried to a depth of 2 - 8 cm. In addition, sediments from WSP were evaluated using 10-d *Hyalella azteca* toxicity tests. Results indicate that AVS affects metal bioavailability in the lentic environment more significantly than in the lotic environment, as there was relatively low AVS concentrations in UCFR sediments. Porewater metal concentrations were less variable than bulk sediment concentrations, and for the UCFR did not exhibit the upstream-downstream concentration gradient typically observed with bulk sediments. For WSP, sediment toxicity was explained by SEM-AVS concentrations or by IWCTU. Direct measurement of sediment porewater metals concentrations, through the use of *in situ* peepers, appears to be a valid approach for evaluating the bioavailability of metal mixtures in freshwater sediments.

114 Biogeochemical Controls on Metal Mixture Bioavailabilities to Meiofauna in Estuarine Sediments. Hagopian, T.A.*; Greiveldinger, S.M.; Chandler, G.T.; Shaw, T.J., University of South Carolina, Columbia, SC. Sediment phase partitioning and metal mixture toxicity (Cd, Cu, Ni, Pb, and Zn) were studied in meiofaunal microcosms from salt marsh sediments. Microcosms consisted of undisturbed cores (f=12 cm) from a pristine salt marsh at North Inlet, SC, USA. A pre-defined population of rare harpacticoid copepods (*Amphiascus tenuiremis*) was also added to microcosms. Metal exposure was accomplished by adding a layer of spiked sediment containing an equitoxic metal mixture to achieve molar ratios of simultaneously extractable metals/acid volatile sulfides (SEM/AVS) of 1, 2, and 10. The equitoxic metal mixture was such that each metal's concentration was 1/5 the 96-h *Amphiascus* LC_{50} in single metal toxicity tests. The control treatment, which received unspiked sediment, and each of the SEM/AVS ratio treatments consisted of four replicate microcosms, one of which served as a chemistry replicate. Microcosms were maintained under flow through conditions in environmental chambers (20|C, 16:8 L:D photoperiod) for 14 days. Water quality parameters were measured every other day and the microcosms were fed twice a week. Metal concentrations (pore water and sediment) and metal binding phases, such as organic carbon (dissolved and total) and iron- and manganese-oxides, were measured in addition to SEM/AVS. Native meiobenthos were analyzed for impact on community composition, and *A. tenuiremis* was analyzed for effects on age-structure, reproduction and population growth. Results emphasize that in the sediment oxic horizon where AVS concentrations were low (0.1mM/g), other binding phases were likely controlling metal bioavailability to meiofauna. For example, cadmium spikes in sterile sediments up to two times the 96-h *Amphiascus* LC_{50} produced little to no toxicity in whole sediment microcosms.

115 The Best of Both Worlds: Improving Sediment Assessment by Combining the Use of Empirically-Derived and Equilibrium Partitioning Approaches. Berry, W.J.*, U.S.EPA, Narragansett, RI; Field, L.J. , Long, E.R., NOAA, Seattle, WA; Hansen, D.J. HydroQual, Mahwah, N.J.; Ingersoll, C.G. USGS, Columbia, MO; Keating, F.J., U.S.EPA, Washington, DC; Mount, D.R., U.S.EPA, Duluth, MN. Two major approaches exist for deriving sediment quality guidelines for metals in sediments. *Empirically-derived* approaches evaluate the associations between measures of biological effects and measured dry weight concentrations of metals in sediments to derive numerical guidelines associated with certain frequencies of effects, e.g. ERMs, PELs, AETs. The *equilibrium partitioning* (EqP) approach uses insights into the sediment phases that control the bioavailability of mixtures of cadmium, copper, lead, nickel, silver, and zinc to assign a causal relationship between the toxicity of metal in sediments and normalized metal concentrations, e.g. SEM-AVS, IWTUs. We will demonstrate, with field data, a framework for using the two approaches together. The empirically-derived approach is useful with typical harbor sediments, which contain other contaminants in addition to metals. However, in sediments where toxicity is the result of metals alone empirically-derived approaches may be overprotective because these approaches do not account for bioavailability. In typical harbor sediments the EqP metals approach can determine if metals concentrations in sediments should not be toxic, and can indicate sediments in which metals are potentially the cause of observed toxicity, but cannot predict toxicity caused by other substances. The two approaches answer fundamentally different questions: *Is this sediment likely to be toxic?* (empirically-derived) and *Could these metals cause toxicity in this sediment, at these concentrations?* (EqP). Therefore, both the goals of a particular assessment and the strengths and limitations of the different approaches must be considered before an approach, or better yet a selected combination of approaches, is applied.

116 Isolating Individual Stressor Effects at Sites with Contaminated Sediments and Waters. Greenberg, M.*, Rowland, C., Burton, G.A., Jr., Wright State University, Dayton, OH; Hickey, C., National Institute of Water and Atmospheric Research, Hamilton, New Zealand; Stubblefield, W., ENSR, Ft. Collins, CO; Clements, W., Colorado State University, Ft. Collins, CO; and Landrum, P., Great Lakes Environmental Research Laboratory, NOAA, Ann Arbor, MI. Most aquatic sites which are degraded are influenced by multiple stressors. The adverse effects on receptors in these ecosystems depends on many factors relating to their exposure and sensitivity. Using an integrated assessment approach, we characterized physicochemical conditions and benthic macroinvertebrate communities, habitat, and conducted both laboratory and field (*in situ*) stressor evaluations. The study sites were primarily contaminated with PCBs, PAHs, metals, and/or ammonia. Exposures were partitioned into overlying water (low and high flow), sediments, and interstitial waters. Contaminant concentrations were monitored in surrounding media and tissues of indigenous and surrogate organisms. Stressors were partitioned via physical and chemical-based manipulations, both in the laboratory and *in situ*. Uncertainty was reduced via extensive field testing and use of an integrated weighting approach. These deterministic approaches were able to define which media and stressors contributed primary effects to test species and, therefore, have widespread applications for environmental assessments.

117 Development and Application of TCDD Toxicity Equivalence-based Sediment Quality Criteria for Protection of Fish. Cook, P.M., Burkhard, L.P., Mount, D.R., U.S. EPA, Duluth, MN. Sediment quality criteria (SQCs) for persistent, bioaccumulative chemicals, such as PAHs, have typically been developed for the protection of benthic invertebrates. Complex mixtures of 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) and structurally related chemicals are highly toxic, through an aryl hydrocarbon receptor (AhR) mediated mechanism of action, to fish and other vertebrates, but not to invertebrates. SQCs for protection of fish from toxic effects of complex mixtures of AhR agonists during early life stage development can be developed from embryo TCDD dose-response relationships. Consistent water quality criteria can be developed in the same manner. Use of the TCDD toxicity equivalence additivity model for application of an AhR agonist mixture-based SQC requires use of TCDD toxicity equivalence factors (TEFs) and biota-sediment accumulation factors (BSAFs) to relate a safe TCDD toxicity equivalence concentration (TEqC) in fish to associated concentrations of individual chemicals in sediment. TEFs based on early life stage mortality in trout are now available for most potent AhR agonists. BSAFs for fish embryos, whether measured or predicted, reflect the non-equilibrium nature of chemical partitioning between sediments and pelagic food chains. This non-equilibrium condition is associated with site-specific differences in the distribution of chemicals between sediment and water and chemical-specific differences in metabolism by fish. Dynamic mass balance models, which predict the steady-state distributions of each chemical from external sources to sediments in the ecosystem, are needed for determination of chemical loading limits which will ensure compliance with a TEq-based SQC.

118 Critical Body Residues: An Approach to Assessing Organic Contaminant Toxicity with the Marine Amphipod *Ampelisca abdita*. Fay, A.A., Brownawell, B., Elskus, A., MCElroy, A., SUNY, Stony Brook, NY. The objectives of this research are to (1) assess the relative intrinsic toxicity of representatives of various compound classes (PAHs, PCBs, alkylphenol ethoxylates) as it relates to lethal body burdens in *Ampelisca abdita* and (2) determine the utility of the critical body residue (CBR) approach in assessing toxicity of sediment associated contaminants to a benthic invertebrate commonly used in toxicity tests. The compounds have been chosen based on their prevalence in contaminated sediments in the environment, their presumed mode of toxic action (narcosis), and their applicability to the CBR approach. In order to determine the relative acute toxicity of the different compound classes, *A. abdita* are being exposed to water and
sediment spiked with a range of concentrations of radiolabelled benzo(a)pyrene, tetrachlorobiphenyl, nonylphenol, and mixtures of these compounds. Standard 4 and 10 day toxicity tests are being conducted and percent mortality determined. Lethal body burdens of these compounds in *A. abdita* are also being determined. According to the CBR model, the acute toxicity of narcotic compounds when the critical molar volume of the compound in the membrane has been reached, and is not dependent on the exposure concentration or medium. These data should resolve whether the acute toxicity observed as a result of various organic contaminants is occurring at body burdens predicted by narcosis, according to the CBR model. This research represents the first application (to our knowledge) of this technique to *A. abdita* and allows the comparison of acute toxicity and lethal body burdens in *A. abdita* resulting from aqueous and spiked sediment exposures. The CBRs obtained will indicate whether or not these compounds are acting via narcosis, and the additivity of effects.

119 Chronic Exposure of Freshwater and Saltwater Invertebrates to Sediment Containing a Mixture of High K_{ow} PAHs. Spehar, R.L.*, Mount, D.R., Lukasewycz, M.T., Mattson, V.R., Leonard, E.N., Burkhard, L.P., U.S. EPA, Duluth, MN; Burgess, R.M., Serbst, J.R., Berry, W.J., U.S. EPA, Narragansett, RI. Virtually all organism exposure to chemicals in the environment occurs from mixtures. However, our understanding of ecosystem responses and risks associated with chemical mixtures in water and sediment is limited. The U.S. EPA is now considering the development of sediment quality criteria (SQC) for total polycyclic aromatic hydrocarbons (PAHs) that would be more protective of aquatic life than SQC for individual PAHs. This requires knowledge of which PAHs should be included in the *total*. Although the toxicity of lower molecular weight PAHs to aquatic organisms has been shown to be additive, most high K_{ow} PAHs and alkylated PAHs are not sufficiently soluble to be acutely toxic. Thus, their importance relative to the toxicity of the mixture is unknown. To determine the contribution of high log K_{ow} PAHs and alkylated PAHs to sediment toxicity, spiked sediment tests were conducted with both freshwater and saltwater invertebrates. *Hyalella azteca* and *Leptocheirus plumulosus* were exposed for 28 days to a mixture of 13 PAHs (log K_{ow} 5.4-6.8) that was spiked into a natural freshwater sediment at four concentrations (100% mixture = ~4.16 toxic units (TU), 50% mixture = ~2.7 TU, 25% mixture = ~1.56 TU and 12.5% mixture = ~0.81 TU) and to control sediment. Concurrently, *Mysidopsis bahia, Ampelisca abdita* and *L. plumulosus* were exposed for 28 days to the 100% mixture in a natural saltwater sediment. Endpoints studied over the test period were survival, growth, reproduction and tissue residues. Test results are interpreted in the context of assessing risk from sediment-associated PAH mixtures in aquatic environments.

120 Predicting the Toxicity of PAH Mixtures in Field Collected Sediments. Di Toro, D. M. HydroQual, Inc., Mahwah, New Jersey. A method based on Equilibrium Partitioning theory and a narcosis model of toxicity for predicting the toxicity of PAH mixtures in field collected sediments is proposed. Several sources of field collected sediment PAH concentration and associated amphipod effects data were compiled. The number of individual PAHs measured in the sources varied from 13 to greater than 40. A subset of 23 commonly measured PAHs was selected and the sum of these PAHs was found to be representative of the total sediment PAH concentration. For data sets where greater than 40 PAHs were measured, the sum of these 23 PAHs was found to be approximately 70 percent of the total. For data sets where less than the 23 PAHs were measured, missing PAH data were computed using relationships between the concentrations of individual PAHs and total PAHs. These relationships were developed using data from the different sources and were found to be highly significant and consistent across sources. Total measured PAH values were normalized to the sum total from 23 PAHs. Dose-response relationships of amphipod mortality versus total PAH sediment concentration suggest that 50 percent mortality is achieved at concentrations of approximately 10 to 100 mmol total PAH/g_{OC}. For field sediments where the causality was known to be from PAHs, these levels were predicted within a factor a two. Similar results were observed from laboratory spiked sediment experiments where toxicity to amphipods from PAH mixtures was investigated. This approach of defining total PAH concentration was useful at locations where only a few low concentration PAHs were measured and significant toxicity was observed because it provided a method of determining whether or not the causality could be attributed to the unmeasured PAHs.

121 The Effects of Multiple Stressors on Estuarine Ecosystems: The Influences of Nutrient Enrichment on Trace Element Cycling. Riedel, G.F.*, Sanders J.G., and Breitburg, D.L. Academy of Natural Sciences, St. Leonard, MD. As part of a larger program (COASTES), we used 1000 L continuous-flow (10% per day turnover) mesocosm experiments to determine the effects of nutrient (N and P) and trace element (As, Cu, Cd, Ni and Zn) additions as interacting stressors on estuarine ecosystems. Nutrient additions of 16 mM N and 1 mM P, and trace metal additions of 10 mg/L As and Zn, 1 mg/L Cd, 5 mg/L Cu and Ni were used, as well as treatments of trophic complexity: copepods (*Acartia tonsa* and *Eurytemora affinis*), fish (*Fundulus heteroclitus*), and sediment with and without benthos (*Crassostrea virginica, Macoma balthica* and *Heteromastus filiformis*). The nutrient additions had striking effects on the cycling of trace elements in the mesocosms. In nutrient enriched mesocosms, dissolved trace element concentrations were substantially lowered as a result of biological and benthic uptake, in some cases greater than 50%. Nutrient enrichment also led to increased transformation of inorganic arsenate to monomethyl and dimethyl As. Furthermore, nutrient enrichment caused concentrations of certain trace elements, notably Cd, to be present in higher concentrations in the phytoplankton. Other than the rather large effect of sediments reducing the dissolved trace element concentrations, the effects of the other trophic treatments on trace element cycling were minor. These results suggest that anthropogenic nutrient enrichment can have significant effects on the retention, speciation and the trophic transfer of trace elements in estuarine and coastal marine ecosystems that depend on the specific behavior of each element.

122 The Effects of Multiple Stressors on Estuarine Ecosystems: Seasonal Variability in Response of Estuarine Phytoplankton Communities. Sanders, J.G.*, Weinstein, M.J., Breitburg, D.L., Academy of Natural Sciences, St. Leonard, MD. As part of a larger program (COASTES), we have examined the response of natural phytoplankton communities from the Patuxent River (a subestuary of Chesapeake Bay) to multiple stressors (nutrients and the trace elements As, Cu, and Cd). A number of replicate runs were performed in 1996 and 1997 in 1 m^3 mesocosms in which phytoplankton were exposed to either ambient or elevated levels of nutrients and trace elements, separately and in combination, over a 5 week period. Phytoplankton biomass, species composition, size distributions, and productivity responded to both stressors, but not in a manner expected from previous mesocosm experiments. Response to trace elements was more variable both temporally and among phytoplankton taxa than was the response to nutrients. Integrative measures of phytoplankton response (biomass, productivity) tended to be reduced by trace element additions. For individual taxa, however, trace elements resulted in a mix of positive and negative responses. The most dominant species, a large centric diatom, *Rhizosolenia fragilissima*, was greatly stimulated by elevated nutrients, and often inhibited by the trace elements. Interactions between the stressors were important in regulating phytoplankton response. In particular, seasonal shifts in the nutrient that limited phytoplankton growth in the Patuxent River (P in spring, N in mid-late summer and fall) greatly affected the toxicity of the trace elements, presumably by either facilitating (when P was limiting) or impeding (when P was abundant) As uptake, incorporation and subsequent toxicity.

123 Anthropogenic Vs. Natural Causes of Hypoxia and Anoxia in the Eutrophying Neuse River Estuary, North Carolina: What Is Manageable and What Is Not? Paerl, H. W.*, Pinckney, J. L., Fear , J. Peierls, B. L. UNC-CH Institute of Marine Sciences, Morehead City, NC. The contrasting impacts of nutrient-stimulated phytoplankton blooms vs. storm-driven organic matter enrichment on oxygen (O_2) depletion were examined and evaluated in the eutrophic, salinity-stratified Neuse River Estuary, North Carolina, USA. This nitrogen (N)-limited estuary is experiencing increasing anthropogenic N loading from accelerating urban, agricultural and industrial development in its watershed. Resultant algal blooms, which provided organic matter loads capable of causing extensive low O_2 (hypoxic) and depleted O_2 (anoxic) conditions, have induced widespread mortality of resident fin- and shellfish. Phytoplankton blooms followed periods of elevated N loading, except during extremely high runoff periods (e.g., hurricanes), when high rates of flushing and reduced water residence times did not allow sufficient time for bloom development. During these periods, hypoxia and anoxia were dominated by watershed-derived organic matter loading. Contrasting organic matter loading scenarios were examined in sequential years (1994-1996) to compare the differential impacts of an average discharge year (10 year mean hydrological conditions) (1994), N-stimulated summer algal blooms (1995), and a major hurricane (Fran, Sept. 1996). The response of primary production, hypoxia and anoxia to these contrasting hydrologic years and resultant organic matter loadings help distinguish anthropogenic from climatic modulation of O_2 dynamics and fish kills.

124 Nitrogen Fixing Cyanobacteria in the Neuse River Estuary: Potential for Growth of Native and Non-native (*Nodularia* Sp.) Bloom Forming Organisms. Moisander, P. H.*, Piehler, M. F., Paerl, H. W., Institute of Marine Sciences, University of North Carolina at Chapel Hill, Morehead City, NC, USA. Global expansion of harmful algal blooms has been related to increased availability of nutrients in coastal and estuarine environments. Reduction in nitrogen loading has been proffered as a means to control nuisance algal growth in the Neuse River Estuary. However, evidence from other freshwater and brackish ecosystems, suggests increasing potential for N_2-fixing cyanobacteria when there is a decrease in the N:P ratio (ratio of dissolved nutrients). We investigated the potential of Neuse water to support growth of two strains of a brackish water, hepatotoxic diazotrophic cyanobacterium *Nodularia*, isolated from the Baltic Sea. When phosphorus was supplied in excess, biomass (chlorophyll *a*), productivity, and N_2-fixation of the cultures grown in the Neuse water were at a similar level as in the growth medium. When the inoculant was conditioned in a reduced, natural PO_3-P concentration (5 mM), biomass increased in the Neuse water only if phosphorus was added. Dilution bioassays were used to assess the potential for native N_2-fixing organisms of the Neuse River Estuary to grow in reduced N:P ratio. In late summer, N_2-fixation by native cyanobacteria (*Anabaena* sp.) was enhanced when N:P ratio was reduced. These results suggest that no chemical inhibition exists for the establishment of *Nodularia* in the Neuse River Estuary, and phosphorus is likely a limiting factor for its continued growth there. Both *Nodularia* experiments and dilution bioassays suggest that reduction in N loading, without restrictions in P loading, may enhance conditions conducive to N_2-fixation by diazotrophic cyanobacteria in the Neuse River Estuary.

125 Spatial Patterns of Chemical Contamination and the Benthic Community Response Across Salt Marshes in Developed Areas. Sanger, D.M.*, University of South Carolina and the SC Marine Resources Research Institute, Charleston SC; Holland, A.F., SC Marine Resources Research Institute, Charleston SC. Defining the relationships between the ecological condition of salt marsh systems and watershed development is a major environmental issue associated with increasing development of the coastal environment. Salt marshes are thought to act as sinks for contaminants; however, it is unknown where these contaminants reside within these systems and the associated benthic community response. Understanding this relationship is critical information for the development of land use plans to prevent degradation of salt marshes and estuaries. In this study, four creeks and their surrounding marsh platform in the Charleston Harbor Estuary were examined to determine the patterns of chemical contamination and the effect on the benthic community. Two tidal creeks/salt marsh systems with developed watersheds were examined along with two reference systems. The two developed watersheds, Diesel and Shem, have industrial and suburban development as the dominant land cover, respectively. Twenty-four sites were examined within each system from the headwaters to the mouth of each creek along transects leading from the creek channel up onto the high marsh. Sediment contaminants measured included trace metals, PAHs, PCBs, and pesticides. The two reference creeks had lower concentrations of contaminants than the developed creeks. The types and patterns of contaminant deposition across the system differed between Diesel and Shem Creeks. The benthic community varied spatially across marsh systems with the dominant organism being a tubificid oligochaete. The intermediate levels of pollution in Shem Creek appeared to cause an enrichment of the dominant oligochaete. By contrast, the higher levels of pollution in Diesel Creek appeared to cause a higher abundance of pollution tolerant polychaetes.

126 The Relationship of Physical, Chemical, and Biological Results of an Ecological Risk Assessment in and Estuarine Environment. Rockett, C.L.*, A. Getty, T. Brzinski, Parsons Engineering Science, Inc., Norcross, GA. Using the EPA's phased ecological risk assessment approach, risks to aquatic organisms were evaluated at a RCRA site adjacent to the Back Bay of Biloxi, MS. This presentation will discuss relationships found amongst the physical, chemical, and biological results and how these relationships affect the interpretation of the risk assessment results. The ecological evaluation was conducted in two phases. The Phase 1 evaluation included sediment and surface water sampling, modeling of exposure to six aquatic receptors representative of several trophic levels, and an estimation of risk using the hazard quotient (HQ). Results of the Phase 1 investigations indicated significant potential risks to these receptors, and a Phase 2 study was conducted. Phase 2 investigations included resampling of sediments at select locations, whole sediment toxicity studies using the amphipod, *Leptocheirus plumulosus*, and the collection of fish tissue for chemical analysis in order to refine the Phase 1 model risk estimations. Results of the Phase 2 investigations indicated risks; however, the magnitude and number of chemicals resulting in risk were greatly reduced. Evaluation of spatial, temporal, and direct interactions of the physical, chemical, and biological conditions indicate that these parameters greatly influence risk assessment results. No single, standardized risk assessment result accurately reflects site conditions and risks to aquatic organisms in this dynamic estuarine ecosystem. Consideration of contaminant transport and sediment parameters as it relates to risk estimates should play an active role in the decision making process for estuarine ecosystems.

127 Duwamish Estuary Ecological Assessment. S. Munger, B. Adamson, S. Mickelson, K. Schock, R. Shuman, J. Simmonds, J. Strand, B. Swarner, L. Wharton, and K. White, King County Department of Natural Resources, J. Toll* and C. Wisdom, Parametrix, Inc. King County has developed a risk assessment framework for answering key questions about the future direction of its combined sewer overflow (CSO) control program for the Duwamish River and Elliott Bay estuary. We evaluated risks to aquatic life, wildlife, and people under existing conditions. We also assessed assuming CSO discharges were eliminated to the system, while all other impacts remained the same as under existing conditions. We will present the results of these risk assessments and discuss how they are being used to support regionally important watershed management decisions. The risk assessment scope was highly ambitious which provided a variety of unique challenges. Foremost among these is the problem of providing a detailed exposure analysis for a wide variety of receptors in a large, diverse, and dynamic estuary. We accomplished this by (1) developing a high resolution, three-dimensional chemical fate and transport model, (2) conducting an intensive field sampling program designed to calibrate the model, (3) conducting additional fieldwork to obtain fish and shellfish tissue concentration data, and information on use of the estuary by people, and (4) using a model post-processing program to extract the exposure information needed to assess risk, from the (for all practical purposes) spatially and temporally continuous simulation results. The use of a sophisticated model provides a great deal of flexibility, which has allowed us to be more responsive to questions from people interested in decisions regarding environmental management in the estuary.

128 An Integrated Fate and Bioaccumulation Model for Polycyclic Aromatic Hydrocarbons in a Marine Ecosystem. Harris, G.E.* and Gobas F.A.P.C. Simon Fraser University, Burnaby, B.C. Canada. Assessing the fate and bioaccumulation of industrial contaminants in the environment is an important component of risk assessment. The sustainable management of chemicals requires defining acceptable environmental concentrations and relating these concentrations to the emissions responsible for the exposure. A time dependent, regional mass balance model using first order kinetics to describe uptake and elimination processes (ECOFATE) has been developed to relate PAH emissions from an aluminum smelter with concentrations in various environmental compartments, including sediments and biota. A large smelter is located at the head of a fjord along the British Columbia coast. Although emission data since the smelter's start-up in 1954 is limited to recent years; sediment cores provide an historical record of contaminant release into the fjord system. This historical profile is combined with our understanding of chemical fate processes to close the mass balance for PAHs in the fjord. The results of further pollution reduction plans by the smelter are presented as environmental concentrations and related to current regulatory criteria. The approach represents a framework for managing emissions of contaminants that meet acceptable concentrations in the environment.

129 Risk Analysis of BTEX in the Estuarine Environment: Estimating Stochastic Exposure from Frequent Low-level Oil Spills to Predict the Consequences. Zach, L.S.* and Keey, R.B. University of Canterbury, Christchurch, New Zealand. The accidental release of chemical contaminants by the process industries can pose both immediate and long-term threats to the natural environment. Long-term damage from frequent, low-level releases is difficult to predict and may not be evident until the damage is done. This paper attempts to analyze potential long-term population problems to an important shellfish bed in a harbor near an oil refinery before they become evident. Such modeling is a relatively inexpensive method, compared with ongoing field-sampling and laboratory analysis, to identify specific chemicals, industrial operations, and sites which might require further testing and investigation. The probable exposure concentration over one operational year from small-scale, oil-spills was estimated from reconstructed historical frequency data and calculations of environmental transport. The exposure concentration of the aromatic fraction of gasoline (a 45% maximum in New Zealand premium grade auto fuel) was estimated involving rate processes such as evaporation, spreading, and dispersion in an estuarine environment. These modeled exposures over time were compared with published sub-lethal, sensitive life-stage and adult lethal marine toxicity criteria. Stochastic results indicate sublethal exposures greater than 10 ppm BTEX at average intervals of 11 days and lethal exposures of over 100 ppm BTEX at average intervals of 25 days. This exposure frequency may not provide enough time for the shellfish to adequately recover. Given the presently available distribution of spillage size and frequency, damage to the long-term health and population of this shellfish bed is possible. Further, more detailed analysis and monitoring are recommended.

130 El-Mex Bay (Alexandria, Egypt) Ecological Assessment. Khan, A.A., EA Engineering, Science and Technology, Sparks, MD, USA. The study was conducted as part of the Mediterranean Sea Environmental Characterization Program (MSECP) which supported an Environmental Assessment (EA) commissioned by the United States Agency for International Development (USAID). The objective of the study was to characterize the existing environment in the bay and predict impacts likely to result from implementation of six effluent disposal alternatives being evaluated as part of the EA. Except for the ocean outfall alternative, all other options envisaged continued discharge of either primary treated wastewater (No Action Alternative) or secondary treated wastewater into the bay. Environmental characterization included monthly analyses of water and sediment samples for a suite of analytes and seasonal assessment of abundance and diversity of biota from representative trophic levels. Results indicated that the bay continues to remain a polluted eutrophic system. Biota analyses revealed that community composition of primary and secondary producers has undergone significant changes in the last decade. Potential impact assessment showed that the bay environment would be positively impacted under the ocean outfall alternative. Positive impacts included improvement in water and sediment quality and decreased eutrophication.

131 Evaluation of Multiple Endpoints Resulting from Divalent Metal Exposures to the Embryo of the Japanese Medaka. M. L. Odom*, J. M. Redding, Tennessee Technological University, Cookeville, TN, and M. S. Greeley, Jr., Oak Ridge National Laboratory, Oak Ridge, TN. Japanese Medaka, *Oryzias latipes*, embryos were exposed to a range of divalent metals including cadmium (Cd), copper (Cu), manganese (Mn), mercury (Hg), nickel (Ni) and zinc (Zn) using an individual culture method. No Observed Effect Concentrations (NOECs) and Lowest Observed Effect Concentrations (LOECs) for hatching success and survivorship ranked as follows Hg < Cu < Cd , Zn , Ni < Mn. Survivorship, hatching success, and incidence of developmental defects were evaluated as measures of effect by the various metals. In the case of mercury, hatching success and developmental defects were more sensitive measures of effect than survivorship. Developmental defects varied by metal. Mercury was the strongest inducer of spinal and optic defects.

132 Site Characterization of the Proposed Nicolet Mine, Crandon, Wisconsin: What Happens if the Swamp is Drained and Copper Enters Via the Runoff? Nimmo, D. R.*, MidContinent Ecological Science Center, BRD/USGS, Ft. Collins, CO; Castle, C. J., Colorado State University, Ft. Collins, CO; Pillsbury, R. W., Environmental Division, Sokaogon Chippewa Community, Crandon WI. The questions raised in the title became the basis of the study plan. We tested resident species such as *Ceriodaphnia dubia, Pimephales promelas*, and after some methods development, we also tested wild rice, *Zizania aquatica*, and walleye, *Stizosdedion vitreum*. The rice and walleye are considered as valued resources by the Sokaogon-Band of the Chippewas. Because we are in the *Site Characterization* phase, we are formulating some questions about how the toxicological data relate to the construction and operation of the mine. Surprisingly, the order of decreasing sensitivity of organisms to copper in natural or reconstituted water using a variety of endpoints are daphnids, wild rice, walleye and fathead minnows. Dissolved organic carbon (DOC) appears to govern the toxicity of copper in Swamp Creek water.

133 Lead Effects on a Brazilian Tropical Fish Assessed by Blood Parameters and Morphological Gill Alterations. Martinez, C.B.R.*, Zaia, C.T.B.V. and Nagae, M.Y. Universidade Estadual de Londrina, Paraná, Brasil. Laboratory toxicity tests were conducted to evaluate the effects of lead on the Brazilian fish *Prochilodus lineatus*. Adult fishes were exposed to two sublethal concentrations of $Pb(NO_3)_2$: 38 and 114 ppm, corresponding to 25 and 75% of $LC_{50(96h)}$ respectively. Gills and blood samples were withdrawn after 6, 12, 24, 48, 72 and 96 hours of lead exposure. The results showed a significant decrease on blood sodium concentration after 48h of exposure to 114 ppm in relation to the control group. A significant increase on blood glucose were observed after 6h of exposure to both lead concentrations followed by a return to the basal values after 48 hours; a significant decrease were observed on blood lipids and cholesterol after 6 hours of exposure to both lead concentrations; a decrease of blood total proteins were observed after 6h of exposure to 114 ppm and after 72 hours to 38 ppm of lead nitrate. Morphological alterations such as lamellar aneurism, epithelial lifting and hyperplasia with lamellar fusion were observed in gills of fishes exposed to lead. Animals exposed to 38 ppm of $Pb(NO_3)_2$ showed 100% incidence of epithelial lifting, in all experimental periods, followed by hyperplasia and lamellar aneurism. In 114 ppm of $Pb(NO_3)_2$ it was observed a major occurrence of hyperplasia and lamellar fusion. These results suggest that sublethal lead concentrations promote a disruption on sodium hyperegulation, a typical stress metabolism response and some morphological alterations on gills that might interfere on gas exchange.

134 Sensitivity to Cadmium and Chromium of a Mexican Wild Population of *Hyalella azteca*. Ramírez-Romero, P. Universidad Autónoma Metropolitana-Iztapalapa, Mexico City, Mexico. The objective of the present study was to evaluate the sensitivity to metals of a wild population of *Hyalella azteca* found in Mexico City. Because our laboratory intends to do research in freshwater ecosystems and *H. azteca* is a standard species in aquatic toxicology a controlled culture of these amphipods was started with organisms found in a local lake. However, since *H. azteca* has been shown to be a genetically complex species, we wanted to compare the relative sensitivity of the Mexican amphipods to the data reported for this species in North America and also, to compare the data with the limits established by the Mexican legislation to learn if they would protect this species. Animals were collected at Lake Huayamilpas, a small lake located in the southern part of Mexico City. The amphipods were cultured in moderately hard reconstituted water at 24± 1 BC and natural photoperiod. Animals were acclimated for 1 week and during the following 2 weeks juveniles were obtained for the toxicity tests. Standard static acute toxicity tests were performed using $CdCl_2$ and $K_2Cr_2O_7$ separately. The calculated 48 h LC_{50} for cadmium and for chromium were 1.77 Fg/L and 170.55 Fg/L respectively. Mexican amphipods were 175 times more sensitive to cadmium than the species average reported by the US EPA and up to 7.9 times more sensitive than the lowest cadmium LC_{50} value published for this species. In contrast, Mexican amphipods were only 3.7 times more sensitive to chromium than the species average reported by the US EPA. From the regulatory point of view these results are a red flag because water quality criteria in Mexico are in the mg/L range.

135 Conducting Laboratory Studies to Evaluate the Site-Specific Toxicity of Cu. Lipton, J., Welsh, P.G., Hagler Bailly Services, Boulder, CO. Laboratory toxicity tests increasingly are focusing on the site-specific toxicity of heavy metals. These tests include bioassays performed with reconstituted laboratory water, with field collected waters, and in competitive bioassays (e.g., *water effects ratio* tests performed in the United States). We evaluated the site-specific toxicity of Cu in a series of laboratory bioassays performed both with field collected water and reconstituted waters. We present results related to methodological considerations necessary to ensure adequate evaluation of toxicity in a field setting. Specifically, we discuss the relative effects of calcium and magnesium on Cu toxicity, pH interactions, accumulation of dissolved organic carbon (DOC) in bioassay aquaria, and influences of water collection and storage in tests using field collected waters. In addition, we present information on characterizing the metal binding affinity and capacity of DOC in field collected waters using geochemical methods (Cu ion specific electrode, ion exchange) and linking field derived metal binding capacities to laboratory derived toxicity test results

136 The Effects of Salinity on Chironomid Cadmium Body Burdens. Barjaktarovic,L.*, and L.I. Bendell-Young, Simon Fraser University, Vancouver, BC, Canada. The Risky creek area of B.C. Canada contains ponds which range in salinity from 0-10 ppt each with established invertebrate communities. Previous studies found no relationship between chironomid cadmium body burdens and the sediment geochemistry of each pond. Hence, we hypothesized the specific adaptations of the sampled chironomid populations which allowed them to survive under different saline regimes also influenced their ability to accumulate trace metals such as cadmium. To test this hypothesis, chironomids were sampled from study ponds and four different cultures were established in the lab. Two cultures were established from a low salinity pond (I, II), one from a mid salinity pond (III) and one from a high salinity pond (IV). As well, a culture of *Chironomus tentans* (V) was used. Fourth instar larvae from each culture were placed in three aquaria containing 0.6L of water of a different salinity(0, 0.8, 4.2 ppt) and 3 microCuries of Cd-109/L. Prior to introduction to aquaria chironomid larvae were acclimated to their test salinity for ten days. The experiment ran for fourteen days after which the organisms were measured for Cd-109 content. Cultures I, II and V showed similar and expected trends in that Cd body burden decreased from low to mid salinity and the chironomids did not survive at high salinities. However, in the low salinity tanks, culture I had significantly lower Cd levels than cultures II and V, as did culture III. Culture III (from the high saline wetland) was unique in that Cd body burdens increased significantly with increasing salinity until, at 4.2 ppt, Cd levels reached that of culture IV, which had consistently high Cd levels at all test salinities. These results indicate chironomids in high salinity environments may be at higher risk for cadmium contamination.

137 Heavy Metal Sensitivity of Antarctic Amphipods: Effects of Low Temperature and Gigantism. Ling, N.*, Department of Biological Sciences, The University of Waikato, Hamilton, New Zealand; Hickey, C.W., National Institute of Water and Atmospheric Research, Hamilton, New Zealand; Burton, G.A., Institute for Environmental Quality, Wright State University, Dayton, OH. The marine environment of Antarctica is characterised by extremely stable thermal and physical conditions, and a surprisingly high incidence of biological gigantism. Very few studies have investigated the sensitivity of polar marine organisms to potential anthropogenic contaminants. Some existing data from acute toxicity tests suggests that these animals may show remarkable insensitivity to some toxicants. This study aimed to evaluate the effect of size and time on the response of necrophagous (flesh eating) amphipods to heavy metals. Laboratory toxicity tests were performed at New Zealand's Scott Base, McMurdo Sound, Antarctica, using different life stages of three species of marine amphipod, *Orchomene plebs*, *Orchomene pinguides*, and *Paramora walkeri.* Tests compared the median lethal concentrations of zinc, copper, chromium, cadmium and lead. Test duration was from 4 to 14 days and test temperature was -1 +/- 0.5°C. Tests were static with replacement and animals were fed every four days. Results indicate that these species are similar in sensitivity to comparable organisms from more temperate latitudes, however the lethality time response is heavily dependent on the low environmental temperature and large size of some of these organisms. Prior estimates of the sensitivity of polar marine organisms that contend heavy metal insensitivity may be biased by short exposure duration. Our results have clear relevance to considerations of pollutant impact in polar environments.

138 Toxicity of Ammonia to Marine Organisms. Boardman, G.D., Starbuck, S.M., He, H.,Virginia Tech, Blacksburg, VA, Hudgins, D.B., Li, X.Y., Olver Incorporated, Blacksburg, VA. Laboratory toxicity tests were performed to obtain more data pertaining to the toxicity of ammonia to saltwater organisms. The standards for instream ammonia limits in marine environments are presently based on toxicity tests involving both freshwater and saltwater organisms. Acute tests (48 hour and 96 hour) were performed at 20 °C, and chronic tests (7 day) were performed at 25 °C. Synthetic water and natural water taken from the Chesapeake Bay were used. The results obtained are given below in terms of mg/L of unionized ammonia. For the Sheepshead Minnow, the 48 hour LC50 and the 96 hour LC50 were 2.68 mg/L and 2.09 mg/L, respectively, and the 7 day NOEC (growth) was 0.34 mg/L. For the Summer Flounder, the 48 and 96 hour LC50's in synthetic water were 1.30 mg/L and 1.08 mg/L, respectively, and the 7 day NOEC (mortality) was 0.11 mg/L. In Chesapeake Bay water, the 48 and 96 hour LC50's were 1.11 mg/L and 0.96 mg/L, respectively. For the Atlantic Silverside, the 48 hour LC50 in synthetic water at salinities 14, 22, and 30 ppt were 1.50 mg/L, 1.17 mg/L and 1.08 mg/L, respectively. The 7 day NOEC (growth) was 0.48 mg/L. In Chesapeake Bay water, the 48 and 96 hour LC50's were 1.45 mg/L and 1.08 mg/L, respectively. For the Mysid Shrimp, the 48 hour LC50 and the 96 hour LC50 were 1.00 mg/L and 0.76 mg/L, respectively. For the Ghost Shrimp, the 48 and 96 hour LC50 were 3.48 mg/L and 1.97 mg/L, respectively, and the 7 day NOEC (growth) was 1.08 mg/L. Based on these results, it appears the standards for instream ammonia limits in marine environments can be somewhat relaxed.

139 Ion-Specific Considerations for the Derivation of Total Dissolved Solids (TDS) Water Quality Benchmarks: A Solvay Process Residue Case Study. Chappie, D.J.*, Fuchsman, P.C., and Barber, T.R., McLaren/Hart, Cleveland, OH. The effects of saline water on freshwater organisms are often assessed based on measurements of total dissolved solids (TDS). TDS represents the total concentration of many ions, the toxicity of which is related to both total osmotic stress and effects of specific ions. Given the variety of potential ion compositions, predictions of toxicity are difficult to assess based on TDS measurements alone. This investigation focused on leachate from Solvay process waste lagoons in northeastern Ohio. The Solvay process was formerly used to produce soda ash from limestone and brine. Based on a literature-reported analysis of Solvay residue leachate composition, an ion-pairing model was used to estimate specific ion concentrations in diluted leachate. Ion compositions were also measured in five ground water samples containing elevated TDS levels and two surface water samples containing low TDS levels. The ion compositions of the ground water samples were generally similar to the modeled Solvay leachate ion composition, but quite different than the surface water ion compositions. The TDS of Solvay leachate was comprised primarily of chloride (50%), while the TDS of surface water samples was comprised primarily of bicarbonate (40%). The toxicity of the measured and modeled ion fingerprints was estimated using the Gas Research Institute Freshwater Salinity Toxicity Relationship (GRI-FW STR) model, which predicts toxicity to three freshwater organisms (*Ceriodaphnia dubia*, *Daphnia magna*, and *Pimephales promelas*). Safe TDS levels were predicted to be higher for Solvay leachate than for the ion fingerprint of uncontaminated surface water, indicating that site-specific water quality criteria for TDS should include consideration of specific ion composition.

140 Hydrogen Peroxide as a Therapeutic Compound for Bacterial Gill Disease in Fish. Tort, M.J.*, G.A. Wooster and P.R. Bowser, Cornell University, Ithaca, NY. Hydrogen peroxide and its primary decomposition products, oxygen and water, are considered environmentally compatible. The apparent lack of adverse environmental impacts of hydrogen peroxide combined with its effectiveness as an antimicrobial agent makes this compound an excellent candidate for further evaluation in aquaculture. Hydrogen peroxide is currently being tested for its potential as a chemotherapeutant for a variety of external disease organisms of fish. Research in our laboratory has focused on delineating effective hydrogen peroxide treatment regimes for treating bacterial gill disease in salmonids (rainbow trout as a model) and walleye. Results from target animal studies indicate that toxicity of H_2O_2 is dependent on species, age, dose and water temperature. Acute toxicity is evident from histopathologic inspection of gills (e.g. extensive epithelial lifting, necrosis) of fish treated with high doses of H_2O_2. The value of hydrogen peroxide as a therapeutic compound for use in walleye culture has been questioned due to the high sensitivity of walleyes to this compound at suggested treatment levels. Studies indicate that exposure of walleye to low levels of hydrogen peroxide would result in increased tolerance to subsequent exposure. This phenomenon of increased tolerance may be of value in the development of management strategies that will permit the use of hydrogen peroxide with walleyes and other sensitive species.

141 Environmentally Acceptable Endpoints for Petroleum Contaminated Soils. W.R. Berti, J.R. Shann, F.C. Hsu, M.V. Johnston, S.D. Cunningham. It is increasingly apparent that the risk that petroleum contaminated soils, sediments, and sludges pose is dramatically affected by the solid matrix, the age of the contamination, and the chemical and biological processes that have occurred over time. The goal of much of our work in this area has been to develop chemical, physical, and biological methods to measure the actual risk posed by these contaminated materials and make predictions concerning clean-up needs and goals. We have shown that supercritical fluid extraction (SFE) and accelerated solvent extraction (ASE) begin to differentiate sequestered compounds from freely available materials. In our research we also have used testing protocols developed for GI tract and dermal uptake of drugs. These methods have proven useful in predicting uptake of toxicants from sludges and soils. The Caco-2 cell permeation (to simulate GI tract uptake) has been measured separately for over 30 radiolabeled compounds (including petroleum hydrocarbons, other industrial pollutants, and pesticides). Dermal permeation measurements of cadaver skin and a silastic sheeting have also been made. A largely similar biphasic correlation between the apparent permeability coefficients (Papp) and log Kow values is evident for both the Cac0-2 cell and dermal permeation experiments.

142 The Impact of Aging on the Aqueous Desorption of Benzene, Toluene, m-Xylene, and Naphthalene from Soil and Comparison with Supercritical CO_2 Desorption. Rixey, W.G.*, University of Houston, Houston, TX; Qu, W., University of Houston, Houston, TX; Garg, S., University of Houston, Houston, TX.; Hawthorne, S., Energy and Environmental Research Center, University of North Dakota, Grand Forks, ND. Research has demonstrated that for organic chemicals that have been exposed to soil for an extended period of time, a fraction of the sorbed chemical exhibits slow rates of desorption. If the rate of desorption is slow relative to other processes such as biodegradation, then the chemical may not have an adverse impact on the environment. To establish more realistic endpoint levels for chemicals in soils, characterization of this rate-limited process is essential. Moreover, more rapid techniques for evaluating the slow rates are desirable, since direct measurements of aqueous release rates can be time consuming and labor intensive. Supercritical CO_2 is one process that could potentially be used for a more rapid asssessment of slow aqueous rates of release. In this research, fixed-bed sorption and desorption experiments were conducted to determine the effect of aging on the rates of both the aqueous and supercritical CO_2 desorption of contaminants for a sandy loam soil and a silty loam soil. Benzene, toluene, m-xylene, and naphthalene were sorbed to the soils from the aqueous phase for varying times (aging) ranging from one week to nine months prior to desorption. Slow desorption characteristics were observed for all four compounds. For example, an aqueous desorption mass transfer rate constant for the slowly released fraction, k_2, of 0.018 day^{-1} was measured for benzene. Desorption was also carried out with supercritical CO_2 at 50, 100, and 150°C and 400 bar for the four chemicals aged for the same aqueous conditions, and desorption characteristics were compared with those for aqueous desorption.

143 In-situ Estuarine Weathering of Sediment-Associated Complex Mixtures: Impact on Water-Extractable Fraction and Toxicity. Hale, R.*, Mainor, T., Harvey, E., Duff, W. Gaylor, M., Powell, D. Virginia Institute of Marine Science, College of William and Mary, Gloucester Point, VA;. Sediment-associated complex mixtures of organic pollutants were allowed to weather in an estuarine environment. Changes in composition and toxicity of the water-extractable fraction (WEF) were examined over time. Toxicity changes were assessed with "Vibrio fisheri", a luminescent bacterium. Fine (high organic) and coarse (low organic) sediments were amended with creosote and two polychlorinated terphenyl formulations (PCT: Aroclor 5432 and 5460) and then placed in jars. Jar arrays were located in sub-, inter- and supratidal areas of an estuarine creek. Each array consisted of eight jars (four containing fine and four containing coarse sediments). Spiked jars were sealed, open, or open but protected from UV.

Open, unspiked jars were also included. Sediments were exposed for 0, 1, 3, 10, 19, 26 and 33 days. After removal, sediments were extracted with estuarine water. Resulting water extracts were filtered, aliquots chemically characterized and tested using Microtox. Twenty-four creosote-derived compounds (including aromatic hydrocarbons, S-, N-, and O-heterocyclics), constituting 90% by weight of the creosote, and the two PCT formulations were tracked. Initially, WEF concentrations were much higher in water derived from coarse sediments. However, creosote-related WEF constituents were present in similar concentrations after one day of field exposure from both sediment types. Measureable concentrations of most constituents remained after 33 days, with the exception of the quinolines. Few degradation products were observed. WEF toxicity was initially greatest from the coarse sediment treatments. After 10 days, toxicities had decreased greatly and were similar between fine and coarse sediment-derived WEFs.

144 Providing Constant Dissolved Concentrations of Hydrophobic Test Compounds by Partitioning from a Solid Phase. Mayer P.*, Wernsing J., De Maagd P.G.D., Tolls J. and Sijm D.T.H.M., Utrecht University, The Netherlands. Toxicity and bioconcentration estimates often rely on the assumption of a constant bioavailable exposure. However, a number of processes can cause dissolved concentrations to decrease to an extent that the interpretation of the results become difficult or even that questions the validity of results. These processes include uptake by the test organism, sorption to glass walls, evaporation and any kind of degradation. We developed a simple method that employs a lipophilic phase to control dissolved concentrations of hydrophobic organics in batch experiments. The test compounds are loaded on a C18 Empore™ disk from where it partitions into the aqueous phase of the batch experiment. This approach, which is named ÒPartitioning Driven Administering (PDA)Ó, resulted in stable aqueous concentrations within minutes to hours for a range of organic compounds (log K_{ow} 3.5 to 7). Aqueous concentrations were proportional with disk loadings spanning a range of up to 3 orders of magnitude, and losses from the aqueous phase were compensated by re-partitioning from the disk, which stabilized the exposure. Consequently, the method allows dosing and maintaining dissolved concentrations of these hydrophobic organics. Validation of the method will be presented together with applications in toxicity and bioconcentration testing.

145 Correlation in the Biological and Chemical Availability of Phenanthrene in Soil During Aging Experiments. Hunter, M. A., Hofstra University, Hempstead, NY, White, J., Nam, K., Alexander, M., Cornell University, Ithaca, NY, Pignatello, J.J., Connecticut Agricultural Experiment Station, New Haven, CT. Hydrophobic organic chemicals sorbed to soil have been observed to be resistant to release and have a low availability to organisms, yet few direct correlations have been made between these two processes. The bioavailability and desorption of phenanthrene aged in two soils (Pahokee peat and Mount Pleasant silt loam) were determined. The rate and extent of desorption, measured under conditions of continuous extraction by Tenax (a polymeric adsorbent), declined with aging. Similarly, the mineralization of phenanthrene by two bacteria and uptake by earthworms declined with aging. Plots of normalized rates and extents of mineralization or desorption as a function of aging period were coincident. The partial removal of organic matter from peat by extraction with dilute NaOH to leave the humin fraction reduced the biodegradation of phenanthrene aged for 38 and 63 days as compared to non-extracted peat, but the effect disappeared at longer incubation times. The rate of desorption from peat previously extracted with NaOH or $Na_4P_2O_7$ declined with aging, and for a given aging period, was significantly slower than from non-extracted peat. Interestingly, the mineralization of aged phenanthrene could be increased by the addition of the non-degradable co-solute pyrene through a competitive displacement mechanism. This work shows that reduction in bioavailability of phenanthrene over time in soil is directly correlated with reduction of its physical availability due to desorption limitations, and that removal of extractable humic substances results in a material in which phenanthrene is even less available. In addition, a nondegradable co-solute will competitively displace aged phenanthrene from soil, thereby increasing physical and biological availability.

146 Sediment Soot Content as a Control on Bioavailability of Hydrophobic Organic Compounds. Lamoureux, E.M.*, Brownawell, B.J. Marine Sciences Research Center, SUNY at Stony Brook, Stony Brook, NY. Soot-amended sediment and un-amended sediment spiked with hydrophobic organic contaminants (HOC) were subjected to laboratory desorption and assimilation efficiency in an effort to assess the importance of soot in controlling HOC bioavailability. The soot was collected from the tailpipes of diesel engine buses and leached with hexane to reduce hydrocarbon concentrations and increase the soot sorption capacity. Three contaminants, naphthalene (NAP), benzo(a)pyrene (BAP), and hexachlorobenzene (HCB) were sorbed to sediments, soot-amended sediments, and soot for a period of 34 days. Simultaneous desorption and *Nereis succinea* assimilation efficiency experiments were then conducted on spiked sediments and soot. NAP and BAP desorption rates for soot-amended sediments (2-4% dry weight) were reduced by at least a factor of two relative to un-amended sediment. Soot, thought to be composed of multi-layered aromatic compounds, might enable introduced polycyclic aromatic compounds (PAH) such as NAP and BAP to establish strong pi-pi interactions and effectively increase the partition coefficient of these contaminants in sediments to which soot has been added. Our results support this hypothesis. HCB desorption rates, however, were similar for both the soot-amended and un-amended sediments. Although HCB is aromatic and planar, similar to PAH, the chlorine substitutions of this molecule may interfere with the formation of pi bonds with the aromatic soot moieties. Preliminary results of *Nereis succinea* assimilation efficiency experiments indicate a trend similar to the desorption experiments; higher assimilation of BAP from un-amended relative to soot-amended sediment and no difference in assimilation between treatments for HCB, although data analysis is still underway. Desorption and *Nereis succinea* assimilation efficiencies of native soot PAH compounds will also be presented and compared with spiked PAH compound results.

147 Bioavailability of Different Forms of Polycyclic Aromatic Hydrocarbons to an Estuarine Amphipod: *Leptocheirus plumulosus.* Merten, A.A.*, Baker, J.E., Chesapeake Biological Laboratory, Solomons, MD, Poster, D., National Institute of Standards and Technology, Gaithersburg, MD. The bioavailability of contaminants stored within, deposited upon, or resuspended from coastal sediments continues to present challenges in assessing the true threat to and potential exposures to marine food webs. We examined the bioavailability of different forms of polycyclic aromatic hydrocarbons (PAHs) to an estuarine, benthic amphipod during a 28 day laboratory experiment. We fed *L. plumulosus* different treatments of PAHs associated w/ *Tahitian isochrysis* (photosynthetic flagellate): 1) labile PAHs (algal cells spiked with perdeuterated PAHs); and, 2) refractory PAHs (SRM 1650 - Diesel Particulate Matter centrifuged with algae) while maintaining constant water and sediment quality, carbon loading (feeding rate), and PAH concentrations between treatments. Field data collected for organic contaminant analysis from the Northern Chesapeake Bay Region do not support Equilibrium Partitioning Theory. Other investigators attribute the deviation from the predicted concentration to the overabundance of combustion-derived (soot-bound) PAHs relative to petrogenically derived PAHs. Petrogenic PAHs may be more readily available to desorb from particulates into lipids than a PAH bound to soot. PAHs produced through high temperature combustion tend to be tightly bound with and absorbed into the soot particle. To test the bioavailability of soot relative to labile PAHs, samples were collected over time (Day 2, 4, 6, 9, 11, 18, 28) to generate uptake curves for both the soot and perdeuterated algae treatments. At each sampling point mortality and growth rate were measured. For Days 18 and 28 reproduction was analyzed and the results between treatments compared. Throughout the testing period, mortality did not significantly differ between treatments, nor did it deviate from the reference treatment (unspiked algae).

148 Surfactant Control of PAH Solubility in *Arenicola marina* Gut Fluids. Voparil, I.M.*; Mayer, L.M., Darling Marine Center, Walpole, ME. The digestive environment is largely responsible for the uptake and subsequent bioaccumulation of polycyclic aromatic hydrocarbons (PAHs) in many marine deposit feeders. We investigated the chemical mechanism responsible for PAH solubilization in gut fluids extracted from a representative deposit feeder, *Arenicola marina.* Surfactant micelle formation led to dramatic increase of phenanthrene and benzo(a)pyrene solubility in gut fluids. Micellar space represented a limited quantity of non-polar solvent for PAH partitioning, and caused enhanced solubilization (compared to seawater solubility) of large PAHs as compared to small PAHs when incubated with contaminated sediments. Incubation at biologically consistent sediment:gut fluid ratios led to saturation of PAH in micelles, thus limiting the amount of PAH digestively available. Repeat incubations of sediment with fresh gut fluid confirmed that PAH release was controlled by the gut fluid chemistry, not the environmental availability of PAHs in the sediment sample. Because surfactants are present in many detritivoresÕ guts, gut passage could strongly influence environmental cycling of PAHs.

149 Fate of Fluorene Compared to Dibenzofuran, Carbazole and Dibenzothiophene in Sediments and Tissues of Finfish. Hellou, J.*, Leonard, J., Antsey, C. Fisheries and Oceans, Nova Scotia, Canada. Meade, J. and Andrews, C. Fisheries and Oceans, Newfoundland, Canada. The fate of lower molecular weight, non-volatile tricyclic aromatic compounds was investigated in biotic and abiotic matrices. The four compounds chosen differed by the presence of a heteroatom in one ring of the linear structures which affected their physical-chemical properties. The biodegradation of the compounds was examined individually using bacterial consortia collected from three marine beaches with differing background chemical histories. Results show differences in the ability of bacterial communities to degrade specific heterocycles, even under optimum environmental conditions. Rainbow trout were also exposed during four months to food pellets spiked with the four polycyclic aromatic compounds (PACs) to compare the relative tissue distribution of the contaminants. Bioaccumulation in muscle, liver, fatty tissue, internal organs and blood vs bioelimination in faeces and gall bladder bile was examined at several time periods. Concentrations were expressed per g tissue, as well as short-term and long-term indicators in reference to total organ load. Results are compared using the log Kow of the contaminants and are used to perform a cautious number of deductions on higher molecular weight PACs. Implications regarding exposure to complex mixtures containing a range of PACs and ensuing biological effects are discussed in terms of knowledge being gained internationally with the hydrocarbon counterparts, i.e. PAHs.

150 An Evaluation of Biological Uptake of Weathered and Dispersed Crude Oil During Mesocosm and Laboratory Experiments with Arabian Light Crude Oil. Coelho, G.M., Wright, D.A., Dawson, R., University of Maryland Center for Environmental Science, Chesapeake Biological Laboratory, Solomons, MD. Aurand, D., Ecosystem Management & Associates, Inc., P0 Box 1199, Purcellville, VA 20134. In April 1998 a mesocosm study was performed to examine the fate and transport of weathered Arabian light crude oil. The experiment involved exposing pelagic, intertidal and benthic organisms to environmentally realistic concentrations of crude oil alone, or in combination with a dipersant Corexit 9500. Six estuarine species were used: sheepshead minnow *(Cyprinodon variegatus),* grass shrimp *(Palaemonetes pugio),* oysters *(Crassostrea virginica),* fiddler crabs *(Uca spp.),* gastropod snails *(Littorina littorea)* and polychaete worms *(Nereis virens).* Animals were sampled at 24, 48 and 96 hour intervals and whole tissue was analyzed for Total Petroleum Hydrocarbons (TPH) using GC-MS. Follow-up laboratory studies were conducted using varying concentrations of the same oil (alone and chemically dispersed) in order to correlate hydrocarbon uptake observed in the mesocosm study with Critical Body Burden (CBB) in selected species. Declining concentration exposures were used in both mesocosm and laboratory studies in order to simulate the dilution that takes occurs after oil is spilled (T ½= 6-12 hours). The results of this type of exposure are compared with LC 50 data found in the literature for similar declining concentration exposures for other organisms.

151 Ecological Risk Assessment and the Precautionary Principle. Fairbrother, A* and Bennett, RS. ecological planning and toxicology, inc., Corvallis, OR. The Precautionary Principle has been touted as an alternative to ecological risk assessment, providing a mechanism for shifting the burden of proof of environmental safety from the general public to the promoter of a particular action. The Precautionary Principle, it has been argued, is based on a holistic approach that views the world as a complex association of interacting parts that can never be completely understood. Risk assessment, on the other hand, traditionally takes a reductionistic approach to understanding cause- and-effects relationships of narrowly defined questions, with the assumption that uncertainty can be reduced through further study. We argue that the risk assessment approach is not incompatible with the Precautionary Principle, only that the scope of risk assessments often is too narrowly defined. The holistic (or "top down") approach frames the question in its broadest ecological and sociological context. Risk assessment and reductionistic scientific principles can then provide a foundation for understanding some (but not all) of the causal linkages between action and effect. The most profound difference in outlook between the two is whether the assessment attempts to minimize the Type (incorrectly assume safety) or the Type II (incorrectly assume effect) experimental error. Both approaches bring to the table biases and beliefs not strictly based on empirical data, arguing for the inclusion of all stakeholders from the beginning, so arguments about model assumptions and uncertainties can take place during the process rather than in an adversarial atmosphere at the conclusion. Combining the comparative risk alternatives and stakeholder involvement aspects of the holistic precautionary approach with knowledge provided by traditional mechanistic scientific studies could result in a more honest dialogue and open information exchange during the environmental management decision-making process.

152 Problem Formulation: Making the Risk Assessment Ecological. Harrass, M.C., Amoco Corporation, Warrenville, IL. Problem formulation is the step in the ecological risk assessment (ERA) process where people decide what valued ecosystem components are to be assessed and what "environmental protection" means for a particular site. Problem formulation is the most distinctive departure from the human risk assessment paradigm and can address three key concerns with ERA: (1) providing a reality check with stakeholders (2) keeping the assessment scaled and focused on the problem, and (3) breaking the barriers that detract from improving ecological risk assessment and management. Reality checks are encouraged by clear statements of endpoints, such as occurred in the assessment of a proposed chemical plant: "Will the walleye fishing in the river be affected?" ERA for other facilities have stumbled when the problem formulation step faltered. Vague endpoints, or late-breaking changes have caused difficulties. Many facilities have, with community assistance, implemented wildlife habitat enhancement programs on facility property, providing a clear identification of valued ecosystem components. "Tiered" assessments attempt to match the scale of the assessment to the scale of the problem. Practical efforts to describe the 'right-sized' ERA are happening in groups such as the ASTM risk-based corrective action (RBCA) program, currently working on standardizing ERA techniques to reflect an appropriate tiering of resource commitment. Barriers to broadened use of ERA have come from several quarters, even from those who wish to further the utility of ERA. These are caricatured by the following statements: "Just give me a table of NOELs" (no-observed effect levels), or "Just tell me what chemicals are/could be present", or "Just follow the checklist." ERA has provided a new and broader opportunity to bring ecological issues to the decision-making table. Problem formulation provides an opportunity to keep the focus on ecology.

153 Unforseen Consequences: Managing Ecological Risks in an Uncertain World. Biddinger, G. R.*, Exxon Company, U.S.A, Houston, TX; and Arnold, W. R., Exxon Biomedical Sciences, Inc., East Millstone, NJ. In September of 1990, the U.S. Government listed the San Francisco Bay Area as a "Toxic Hot Spot" under the Clean Water Act. This action by the U.S. EPA to regulate the San Francisco Bay area led to a series of technical policy decisions which from the perspective of an individual facility resulted in negative environmental consequences. The absence of a formalized methodology for evaluating the net environmental benefits of the site-specific risk management decision was a key barrier to effectively anticipating the environmental consequences of the management strategy. This paper reviews the scientific "weight-of-evidence" underlying the projected environmental risk and explores how conservative technical policy decisions can lead to less than optimum investments in environmental quality. The paper reviews the scientific and technical information within the context of current approaches to assessing environmental consequences. In conclusion, recommendations are provided for developing a formalized methodology to evaluate the net environmental consequences of risk management strategies.

154 Ecological Risk Assessment. Moore, D.R.J.*, The Cadmus Group, Ottawa, ON. A recent paper by Mike Power and Lynn McCarty (ES&T 31: 370-375) noted three myths associated with the practice of ecological risk assessment: a most sensitive species can be selected and appropriately used; chronic data are better suited to regulatory needs than are acute data; and controlled experimental data can be accurately extrapolated to the field. To this list I might add: a safety factor can be selected to account for uncertainties; and a quotient is an indicator of risk. These and any other myths one can dream up, however, only serve as criticisms of the way ecological risk assessment is often practiced, <u>not</u> the underlying scientific foundation. In recent years, we have witnessed a remarkable number of advances in the field of ecological risk assessment and related sciences. Methods are now available to quantify uncertainty and incorporate ecological considerations when estimating risks of single and multiple stressors. Well designed field studies may be used to calibrate the extrapolation from laboratory experiments to the field. Parallel advances have also taken place in the use of ecological risk assessment in environmental decision making. In this paper, I argue that quantifying uncertainty is an essential feature of establishing scientific credibility, discuss how uncertainty may be quantified, and show how such quantification can improve environmental decision making. Other papers in this session will discuss improvements in other facets of the science of ecological risk assessment and its use in decision making. Together, these papers demonstrate that ecological risk assessment, while not yet a mature discipline, is a scientifically valid tool that may be used to help ensure cost-effective environmental decision making.

155 Application and Significance of Nonequilibrium Ecological Dynamics in Ecological Risk Assessment. Landis, W.G.*, Institute of Environmental Toxicology and Chemistry, Huxley College, Western Washington University, Bellingham, WA. It has become increasingly recognized that ecological systems are nonequilibrial in nature as well as highly complex. They are the current product of historical events and, in the context of contaminant exposure, the result of community conditioning. Spatial and temporal dynamics contribute to system nonequilibrium. Differences in the order of original colonization, patchiness, patch orientation, nutrient ratios, and the number of species all can have dramatic influences in mitigating the impacts of contamination. Model simulations demonstrate that effects can range from no apparent impacts to extinction depending upon the spatial and demographic relationships of the patch structure. Multiple stressors may also interact by altering these kinds of landscape arrangements and dynamics. By taking these factors into account it will be possible to conduct ecological risk assessment at the landscape level with important implications for the management of ecological systems.

156 Communicating Science: Incorporating the Output of Ecological Risk Assessments into Environmental Decision-making. Glicken, J. ecological planning and toxicology, inc., Albuquerque, NM. An ecological risk assessment yields specific kinds of scientific data that are intended to be used by decision-makers that have various levels of scientific literacy and who have to make decisions of a wide variety of types, ranging from social policy to business-based. The quality of the communication of scientific information to non-scientists will affect the effectiveness of its use in policy and business decisions. This discussion will present a basic communications model, and use it to identify some of the pitfalls inherent in this type of a 'translation.' It will use examples from existing and historic environmental decision-making processes and debates to illustrate pitfalls and suggest solutions. Points to be addressed will include the encoding of the message, audience identification, and the selection of appropriate transmission vehicles.

157 Using the Relationship Between Multiple Stressors and Valued Resources to Develop a Gis-based Decision-making Tool for Comparative Ecological Risk Assessment. Duncan, P.B.*, Abel, G.A., Bogue, M.W., U.S. EPA, Region 10, Seattle, WA. Using approximately 15 available GIS data coverages for the State of Washington, we examined the relationship between indicators of potential ecosystem stress and valued resources. Potential stressors included point sources of contaminant releases, land use activities such as agriculture, and number of stream segments not meeting beneficial uses. Valued resources included wetlands, priority habitats and species, and number of miles of salmonid habitat. The data were divided among some 1000 hydrologic units across the state. Stressors and resources were combined separately using a simple weighting scheme that also normalized to area. The resulting scatterplot of resources as a function of stress was imposed onto a decision-making framework developed *a priori*. This framework was based on the proposed principle that resources should decline as stress increases and was designed to identify areas that a manager could classify into several groups, namely, (1) act immediately (areas with high values for both stress and resources); (2) watch (areas with low stress and high resources); (3) protect (areas with some stress and moderate resources); (4) restore (areas with high stress and low resources); and (5) obtain information (areas where little seems known about one or both of the indicators). Not surprisingly, the scatterplot demonstrated the lack of complete coverage of information across the state. Similarly, the resulting "management decision" maps indicated where information was lacking and also showed how our estimate of stress was associated with the major north-south interstate corridor. Key improvements could include: accounting for missing data, linking stressors more meaningfully to potential effects, and identifying broad landscape characteristics (ecoregions, for example) useful for stratifying the coverage data.

158 Application of a Food Web-based, Probabilistic Ecological Assessment Tool. Brandt, C.A., Bunn, A.L.*, Eslinger, P.W., Miley, T.B., Pelton, M.A., Pacific Northwest National Laboratory, Richland, WA. A food web-based, probabilistic tool was recently developed which evaluates both radiological and non-radiological doses to aquatic and terrestrial organisms. The food-web architecture is easily adaptable to any site and can be used to support regulatory compliance activities. The terrestrial portion of the exposure model estimates wildlife exposure to contaminants in air through inhalation, in water through dermal exposure and ingestion, and in foods. The aquatic portion estimates exposures to contaminants in surface water and pore water via gill or respiratory uptake, in sediment via dermal exposure and ingestion, and in foods. For terrestrial receptors, model output consists of equilibrium doses for ingestion, inhalation, and dermal routes, total radiological dose, and tissue concentrations. Effective water concentration and equilibrium tissue concentrations are the outputs for aquatic species. All outputs are available as either deterministic values or as statistical distributions generated using a Monte Carlo simulation technique. The tool was used to assess exposures for 56 species to 28 contaminants at 27 locations along the Columbia River at the Hanford Site in southeast Washington state in support of the U. S. Department of Energy's effort to make site cleanup decisions. The species represented all trophic levels for both aquatic and terrestrial environments. The contaminants included radionuclides, metals, and organics. For terrestrial species, contaminants pose potential hazards to some plants, herbivores, and omnivores consuming riverine organisms. The highest effects were due to Cobalt-60, chromium, Cesium-137, mercury, lead, zinc, and Technetium-99 in pore water and sediment. For aquatic species, the organisms most at risk are benthic species. The highest effects were due to cyanide, chromium, copper, mercury, lead, and zinc in pore water and sediment.

159 The Development of Cumulative Effects Assessment Tools Using Fish Populations. Munkittrick, K.R., McMaster, M.E., Gibbons, W.N., Environment Canada, Burlington, ON; Van Der Kraak, G., Dept. Zoology, University of Guelph, Guelph, ON Canada. Until recently, environmental assessments were conducted without regard to either upstream or downstream developments, such that the relative contributions of individual developments could not be separated. There is now (in Canada) a legislated requirement and an expectation that developments will address the cumulative effects of existing developments, prior to the development of methodology for conducting these assessments. We initiated a multi-agency, multi-million dollar, long term study in 1991 which is attempting to address the cumulative aquatic effects of hydroelectric facilities and pulp mill discharges on fish populations within the Moose River basin in Northern Ontario. The Moose River represents a large (>100,000 km^2) sparsely populated (<100,000 people) basin with enormous potential for future industrial development. The focus of our studies is directed from the top-down through interpretation of the impact of alterations in habitat and fish energetics associated with damage at the fish population level, and not on detailed initial characterization of stressors. This approach allows follow-up studies to focus on the critical areas associated with responses of surviving fish within the watersheds, in an attempt to identify limiting environmental conditions and the responsible stressors. The objectives of the project are to provide an interpretation framework using traditional fisheries information to assess the integrated effects of all stressors on the populations at risk, and to isolate key areas and critical stages in life history associated with those changes in fish populations. Over the period of 1991-1998, 50 separate fish collections have been conducted, and diverse issues such as reference site selection, study design and the determination of cause and effect are being addressed.

160 Will the Real Ecological Risk Assessment Please Stand Up? Barnthouse, L. W., LWB Environmental Services, Inc., Oak Ridge, TN. Ecological risk assessment, like risk assessment in general, has been defined in many different ways by many different people. To some, it's a probabilistic modeling technique. To others, it's an immoral substitute for ethical judgement. This paper is premised on a broad definition of risk assessment as a structured process for using scientific information to make rational decisions in an uncertain world. Risk assessment cannot be reduced to a set of standard data types or computational procedures. Moreover, because it is tied to value-laden decisions, the risk assessment process cannot be completely separated from the values of the people or groups making the decisions. The paper dispels some of the fallacies and myths that that follow from inappropriate definitions of ecological risk assessment and discusses three significant challenges to the future of the field: (1) developing rigorous quality assurance standards for both assessments and assessors, (2) ensuring that advances in the scientific disciplines underlying ecological risk assessment are continuously incorporated into assessment practices, and (3) broadening participation in the process by stakeholders who are currently underrepresented.

161 Membrane Lipid Alterations: Exposure to Narcotic Chemicals. Bearden, A.P.*, Sinks, G.D., and Schultz, T.W. The University of Tennessee, Knoxville, TN. *Tetrahymena pyriformis* has been shown to acclimate (i.e., phenotypic change) to the presence of sublethal levels of hydrophobic organic chemicals considered to act via nonpolar narcosis mode of toxic action. The theoretical site of action for narcosis is the cellular membrane. Objective of this work was to parallel population growth kinetics with molecular toxicology to investigate population acclimation to sublethal concentrations of individual chemicals. Population growth experiments were performed in tandem with quantitative molecular analysis of fatty acid content of the pellicle layer of *Tetrahymena*. A control, solvent control, and three sublethal concentrations of model chemical, 1-octanol, were tested. An increase was observed in the amount of fatty acid methyl esters (FAME) for 16:0 and 18:0 (fatty acid notation is the following--carbon number of acyl chain: # of π bonds) with exposure to 1-octanol. Conversely, there was a decrease in the amount of the following FAME: 16:1, 18:1, and 18:2. The overall decrease in the number of π bonds is related a net decrease in overall fluidity. Additionally, there was an increase in the ratio of 16/18 carbon FAME implicating physical accommodation of the compound within the membrane. Molecular changes have been directly correlated with the population growth trends where a concentration dependent lag phase was observed followed by growth rates similar to control populations.

162 Use of Mitocondrial, Photosynthetic and Molecular Endpoints for Risk Assessment of PAHs and PAH/Metal Mixtures. C.L. Duxbury, T.S. Babu, S. Tripuranthkan, D.G. Dixon, B.M. Greenberg, Dept. of Biol., Univ. of Waterloo, Waterloo, ON. Many contaminants, such as PAHs and metals, occur in the environment as complex mixtures, and it is necessary to determine the main contributors to the toxicity. We have been addressing this problem at a mechanistic level by developing rapid bioindicators of effect using PAHs, photomodified PAHs (oxyPAHs), and metals as toxicants. Photomodification of PAHs is widespread, and results in very complex mixtures. We have determined the toxicity of several specific oxyPAHs and examined their mechanisms of impact. Whole organism toxicity of oxyPAHs was assessed with Lemna gibba and Daphnia magna, and *in vitro* with isolated rat liver mitochondria. Most of the oxyPAHs have been found to be toxic, and many are more toxic than their respective parent compounds. The mechanism of toxicity of many oxyPAHs was shown to involve inhibition of electron transport, both in mitochondria and chloroplasts. The specific site of inhibition for selected oxyPAHs was found to be the intermediate cytochromes in the electron transport chains; cytochrome b/c in mitochondria and cytochrome b/f in chloroplasts. Once these cytochromes are inhibited, the quinone electron carrier pools reside in an unstable, highly reduced state. Interestingly, treatment with oxyPAHs caused an increased sensitivity to Cu++ toxicity. This is likely due to metal catalyzed electron transport from the reduced quinone electron carrier pools to O2. As well, some of the oxyPAHs (especially quinones) were found to be electron acceptors, uncoupling electron transport from oxidative phosphorylation. Finally, we are further assessing the mechanisms of PAH impacts at the molecular level by examining gene expression induced by redox imbalance caused by oxyPAHs.

163 Metal Specific Effects on Early Cellular Activation Events. Burnett, K.G.*, MacDougal, K.C. Medical University of South Carolina, Charleston, SC. Binding antigen rapidly triggers early activation events which determine whether a lymphocyte will proliferate, arrest or die. These events include (1) calcium influx (2) tyrosine phosphorylation or (3) activation of one or more extracellular-related kinases (ERKs). *in vitro* exposure to mercuric chloride ($HgCl_2$) and methylmercuric chloride (CH_3HgCl_2) can alter some of these events in peripheral blood leukocytes of a teleost fish, the red drum. The present study compared the effects of mercury with several other metals at a single dose (100 mM) in the same experimental system. Even at relatively low doses (0.1-1.0 mM) $HgCl_2$ and (0.01 - 0.1 mM) CH_3HgCl_2 increased intracellular Ca in fura-2 loaded cells. In the present study uranium and chromium also perturbed Ca flux, while aluminum, lead, cadmium, zinc, nickel, and copper had no effect. Tyrosine phosphorylation was monitored by Western blot with anti-phosphotyrosine antibody and became evident within 1-5 minutes after exposure to $HgCl_2$ (>0.1 mM) or CH_3HgCl_2 (>1.0 mM). None of the other tested metals induced tyrosine phosphorylation. In Western blot with anti-active ERK (Promega), neither $HgCl_2$ nor CH_3HgCl_2 activated the 42 or 44kDa ERKs linked to cell proliferation. In contrast, chromium, lead, aluminum and copper induced activation of these ERKs within 15 minutes. Uranium generated a novel size-form protein which reacted with anti-active ERK. Cadmium, zinc and nickel had no effect on ERK activation. Ongoing studies aim to understand how cells respond to combinations of metals with similar or dissimilar effects on early activation events. [Supported by the Institute for Combined Fisheries Research (FISHTEC)]

164 Copper Mediates Rapid Chloride/hydroxyl Exchange Across Cell Membrane: a Study Using Lamprey Erythrocyte as a Model. Bogdanova A.Yu.* and Nikinmaa M., University of Turku, Turku, Finland,Virkki L.V. University of Yale, New Haven, USA, Gusev G.P., Sechenov Institute, St.Petersburg, Russia. Exposure of fresh water fish to copper causes disturbances in osmoregulation. Molecular mechanisms of copper action on ion and water transport in fish cells and tissues remain poorly understood. We have used lamprey erythrocyte as a model in studying the effects of copper on cell volume, pH and ion transport. Ion fluxes and pH were measured using radioactive tracers and flame photometry. Prelytic CuSO4 concentration of 100 μM caused intracellular acidification and increase in Cl^- influx after a lag period of approximately 2min. The magnitude of copper effect was dependent on extracellular anion decreasing in the series $I^- > Br^- > Cl^- > NO_3^-$. In the presence of ascorbate copper effect was potentiated and there was no lag phase in the response. Pretreatment of the cells with N-phenyl maleimide abolished the copper-induced changes completely. The cellular acid load was followed by activation of amiloride-sensitive Na^+/H^+ exchanger, leading to net Na^+ uptake and cell swelling. Cell swelling activated a regulatory volume decrease mechanism utilizing conductive K^+ and Cl^- pathways. We suggest that cupric ions undergo transformation to cuprous ions on the membrane surface after binding to externally facing thiol group. The poorly water-soluble ion pairs CuCl (CuBr, CuI, CuOH) are formed, penetrate membrane lipid bilayer and carry out electroneutral chloride/hydroxyl ion exchange. The mechanism of copper action resembles the one described before for tributyltin and results in dissipation of pH gradient in all cells with low permeability of membrane to proton equivalents.

165 The Effects of Glutathione Depletion on Copper Cytotoxicity in Oysters, *Crassostrea virginica*. Conners, D.E.*, Medical University of South Carolina, Charleston, SC; Ringwood, A.H., SCDNR/Marine Resources Research Institute, Charleston, SC. Glutathione is a tripeptide that is believed to play an important role in ameliorating metal toxicity. Depletion of glutathione has been associated with an increased risk of metal toxicity in mammals. An understanding of the toxicological significance of glutathione depletion in oysters would be of considerable importance given the widespread use of bivalves in biological monitoring. Laboratory studies were performed using an inhibitor of glutathione synthesis (buthionine sulfoximine) to investigate the effects of glutathione depletion on metallothionein expression, lysosomal membrane destabilization, and lipid peroxidation. In oysters exposred to Cu (20 and 80 $\mu g\ l^{-1}$) and buthionine sulfoximine (20 mg l^{-1}), metallothionein responses were diminished and cellular stress reponses were higher than those observed in oysters exposed singly to Cu. Together, these studies suggest that environmental conditions which cause glutathione depletion may increase the potential for adverse effects during environmental pollutant exposures.

166 Oxidative Stress and the Role of Cytochrome P450 on Pah Phototoxicity: an *in Vitro* Study Using a Fish Cell Line. Choi, J.*, Oris, J.T. Miami University, Oxford. OH. 45056. Many polycyclic aromatic hydrocarbons (PAH) are acutely toxic to aquatic organisms in the presence of solar ultraviolet radiation (SUVR). Our previous studies indicated that oxygen radical related oxidative stress is an important factor in PAH phototoxicity (superoxide anion production and lipid peroxidation product). Oxygen radicals ($O_2^{\cdot-}$, H_2O_2, $OH^{\cdot}$) are very reactive and cause cellular damage through uncontrolled oxidation/reduction of macromolecules such as lipid membrane, DNA and protein. In this study the role of the cytochrome P450 enzyme system (cyt P450) in cellular production of oxygen radicals in PAH/SUVR exposed fish cell line was investigated. Two possible routes of oxygen radical production in PAH phototoxicity were hypothesized. One was that photosensitized PAH molecules directly transfer electrons to O_2 producing oxygen radicals. PAH molecules which lost electrons non-enzymatically may also be involved in redox cycling with concomitant production of oxygen radicals. The other route was that photosensitized PAH accelerate cyt P450 related reactions in which a significant amount of electron leakage can occur. To test this hypothesis topminnow hepatoma cells (PLHC, *Poeciliopsis lucida)* were treated with a cyt P450 inhibitor (0.5mM of clotrimazole) for 3 hours and then simultaneously exposed to a model PAH (anthracene) and SUVR for 2 hours. UV-A intensity was 77 $\mu W/cm^2$. Cell mortality was measured using the MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) assay. At all anthracene concentration levels (350ug/l - 7700ug/L) Clotrimazole treatment increased PLHC cell mortality. This observation implies that direct photosensitization of PAH compound is more important in the production of oxygen radicals and that the cytochrome P450 system functions to reduce the production of oxygen radicals by enzymatic metabolism of PAH compounds.

167 Isolation of a Flavin-Containing Monooxygenase FMO1 cDNA Sequence from the Teleost Rainbow Trout (*Oncorhynchus mykiss*). El-Alfy*, A.T. and Schlenk, D. University of Mississippi, University, MS 38677, U.S.A. Flavin-containing monooxygenases (FMOs)are microsomal enzymes that play a significant role in the biotransforamation and toxicity of thioether pesticides such as aldicarb. In an attempt to determine whether the teleost, *Oncorhynchus mykiss*, expressed FMOs, total RNA isolated from rainbow trout (*Oncorhynchus mykiss*) liver was subjected to reverse transcription-polymerase chain reaction (RT-PCR) using degenerate primers derived from conserved regions of FMO1 and FMO3 proteins. PCR amplification resulted in a single electrophoretic band of 842 bp. The amino acid sequence deduced from this fragment demonstrated closest identity to hepatic rabbit FMO1 (96 %) and pig FMO1 (85 %). Phylogenetic trees were constructed using distance matrix method. These results indicate that the sequence obtained from *O. mykiss* represents a teleost fish *fmo1* gene and is the first identification of an *fmo* gene in a non-mammalian vertebrate.

168 Dioxin Resistance in Killifish (*Fundulus heteroclitus*) from the New Bedford Harbor Superfund Site: *in vivo* and *in vitro* studies of CYP1A1 Inducibility. Bello. S.B.*, Franks, D.G., Stegeman, J.J., and Hahn, M.E., Woods Hole Oceanographic Institution, Woods Hole, MA. New Bedford Harbor (NBH), MA, is a federal Superfund site heavily contaminated with halogenated aromatic hydrocarbons (HAH) including some potent Ah receptor (AhR) agonists. To determine if *F. heteroclitus* inhabiting NBH have developed resistance to AhR agonists, we examined the inducibility of cytochrome P4501A1 (CYP1A1) in fish from NBH and from a reference site, Scorton Creek (SC). The responsiveness of SC and NBH fish was compared following treatment with a model AhR agonist, 2,3,7,8-tetrachlorodibenzofuran (TCDF). The SC fish showed strong induction of CYP1A1 RNA, protein, and activity, in all tissues examined (liver, heart, gut, gill, kidney, spleen and gonad). In contrast, NBH fish showed little or no CYP1A1 induction by any measure, in any tissue. To compare CYP1A1 inducibility by halogenated versus non-halogenated AhR agonists in these fish, EROD activity was measured in primary cultures of *Fundulus* hepatocytes treated with 2,3,7,8-tetrachlorodibenzodioxin (TCDD) or β-naphthoflavone (BNF). Hepatocytes from NBH fish (EC50: 0.604nM) were 14-fold less sensitive to TCDD than SC fish (EC50: 0.043nM), but only 3-fold less sensitive to BNF(NBH EC50:350nM; SC EC50: 113nM). These results indicate that chronic exposure to high levels of HAH has altered the response of NBH *F. heteroclitus* to AhR agonists systemically. The alteration in sensitivity occurs pre-translationally, suggesting a transcriptional effect. It is not limited to halogenated AhR agonists, although the magnitude of the decreased sensitivity is less for non-halogenated AhR agonists. Together these results suggest an alteration in the AhR signal transduction pathway in the dioxin -resistant NBH fish; this is the subject of continuing research. (Supported by Superfund Basic Research Program ES07381 and EPA R823889.)

169 Aryl Hydrocarbon Signaling Components in *Fundulus heteroclitus*: A Phylogenetic Perspective. Powell, W.H.*, Karchner, S.I., Hahn, M.E. Woods Hole Oceanographic Institution, Woods Hole, MA. *Fundulus heteroclitus* is a teleost widely distributed along the Atlantic coast. Abundant in both pristine and polluted estuaries, this species represents a unique tool for the observation of aryl hydrocarbon effects at the ecosystem, population, and organismal levels. To develop this model, we isolated DNA sequences for several components of the aryl hydrocarbon signaling pathway, including two Ah receptor (AHR) cDNAs and an ARNT cDNA. *Fundulus* expresses two distinct AHR genes (AHR1 and AHR2), a phenomenon we have observed in additional fish species. AHR1 most closely resembles the well-characterized mammalian AHRs, but it is more difficult to detect and may not represent the predominant AHR in *Fundulus*. In most fish for which a single AHR sequence is known, the sequence is an AHR2 homolog. The ARNT cDNA isolated from *Fundulus* liver is a homolog of rodent ARNT2. Unlike rodent ARNT2, which displays very restricted expression in adults and embryos, *Fundulus* ARNT can be detected in multiple tissues. Unexpectedly, it resembles both mammalian ARNTs more closely than it does the ARNTb sequence from rainbow trout. Thus, the trout ARNTs represent a third phylogenetic branch that may relate to the tetraploid ancestry of this species. Phylogenetic comparisons reveal remarkable complexity and diversity in the components of aryl hydrocarbon signal transduction. Potentially important differences exist in the number and sequence conservation of these proteins between fish and mammals and among fish species. These differences may contribute to interspecific variations in TCDD sensitivity. Their functional characterization will influence the evaluation of toxicological data from aquatic models, the choice of models for specific studies, and the extrapolation of findings between species. [NIH ES05800, ES06272, ES07381, Donaldson Charitable Trust, WHOI Postdoctoral Scholar Program]

170 Xenobiotic- and Disease-induced Nitric Oxide Synthase: Sequencing the Inos Gene in Channel Catfish, Ictalurus Punctatus, and Designing Diagnostic Antibodies. Rice, C.D.*, Yuan, X. CEHS, College of Veterinary Medicine, Mississippi State University, P.O. Box 9825, Mississippi State, MS 39762, USA. A number of infectious diseases and exposure to some xenobiotics lead to intense necrotic inflammatory responses in fish. While macrophages, other phagocytes, and endothelia are linked to these lesions in mammals, their role in similar lesions in fish is unclear. Macrophage aggregates (MAs) are commonly used as a biomarker of exposure to heavily polluted environments. With regards to infectious diseases, we have recently demonstrated that full-sib families of channel catfish resistant to Enteric Septicemia of Catfish (ESC), caused by the bacteria Edwardsiella ictaluri, have more and larger splenic MAs than do sensitive families. Along with a suite of cytokines and peptides, one of the most potent products of activated macrophages is nitric oxide (NO), a by-product of inducible nitric oxide synthase (iNOS). Expression of iNOS by these MAs, and other tissues, may be an early feature of inflammation and exposure to at least some xenobiotics. Using semi-nested RT-PCR, RACE, and by screening a cDNA library developed from lymphoid tissues of ESC-infected catfish that were also exposed to cadmium and aroclor 1254, we have obtained two overlapping 500 base sequences showing a high degree of homology with both mammalian and rainbow trout iNOS genes. Molecular probes were then developed for northern blotting detection of iNOS expression. In an attempt to develop antibodies to hydrophobic and hydrophilic peptides designed from iNOS sequences expressed from the above library, we obtained a monoclonal antibody that, in turn, recognizes iNOS from the library and in activated leukocytes. These probes may prove to be sensitive early markers for iNOS in infectious diseases and chemically-induced MAs of fish.

171 Development of Soil Cleanup Standards for the Biological Treatment of Wood Preserving Wastes. Jerger, Douglas E., OHM Remediation Services Corp., Findlay, OH. The primary goal of hazardous waste site remediation is to achieve a set of conditions that are environmentally safe for organismal receptors, and will prevent any further action on site. Establishing these conditions raises the issue of "how clean is clean" or "what concentration of a contaminant in soil is environmentally acceptable" for contaminated sites. This debate is occurring on a national scale as part of the Superfund reauthorization. Historically, cleanup goals have not explicitly addressed whether the remedial action results in a safe site. Treatment criteria for soil contaminants have been based on limits of detection (LODs), background concentrations or technical capabilities (Best Demonstrated Available Technology or BDAT). Selecting an appropriate value to represent background concentrations can be problematic in that there is a substantial amount of variability in the natural chemical composition of soils, and many chemicals may have both natural and anthropogenic sources (1). The use of Resource Conservation and Recover Act (RCRA), BDAT, or LOD as a cleanup criteria may also be inappropriate if the remediation goal is to protect human health and the environment. BDAT-based criteria may be over- or under-protective of human health and the environment preventing the use of a more cost-effective treatment technology such a bioremediation. BDAT-based treatment criteria may also focus on a subset of chemicals of limited toxicity, making it difficult to determine if the remedial action removed the health threat posed by other chemicals in the mixture. OHM Remediation Service Corp. (OHM) has recently completed the successful treatment of 14,000 tons of creosote-contaminated material at the Southeastern Wood Superfund Site in Canton, MS. Slurry phase biological treatment was the technology chosen to remediate the contaminated material classified as RCRA KOOl criteria for CERCLA (Superfund) actions: protection of public health, welfare, and the environment.

172 PAH Desorption in a Contaminated Sediment Following Bioremediation and Phytoremediation. Kreitinger. J. P.*, RETEC, Ithaca, NY; Liste, H.-H., Eustis, L., Alexander, M., Cornell University, Ithaca, NY; Neuhauser, E. F., Niagara Mohawk Power Corporation, Syracuse, NY; Hayes, T. E., Gas Research Institute, Chicago, IL. The desorption kinetics for PAHs were determined in a coal-tar and petroleum impacted sediment which had been bioremediated using a tank-based bioslurry treatment system and subsequent phytoremediation. Desorption kinetics were measured using aqueous suspensions of sediment with a C18 (octyl-bonded silica) solid phase sorbent to collect released PAHs. Phytoremediation of contaminated sediment, which had been previously treated as part of an EPA SITE demonstration project, was evaluated using six willow clones, two legume and two grass species in a greenhouse test over an eight month period. Several plant species enhanced sediment PAH losses, including higher molecular weight PAHs, compared to unplanted fertilized control treatments. The distribution of PAHs following phytoremediation was different between willow clones and between plant species. The rapidly released fraction of both lower and higher molecular weight PAHs was greatly reduced following biological treatment.

173 The Role of Intrinsic Bioremediation in Aquifer Cleanup. Rifai, H. S., University of Houston, Houston, Texas, Natural attenuation or intrinsic bioremediation has recently emerged as a viable remediation alternative for many sites contaminated with fuel hydrocarbons and chlorinated solvents that pose little threat to human health and the environment. A number of field protocols, standards and models have been developed to assist researchers and practitioners in implementing intrinsic remediation strategies for their sites. This paper presents the advantages and limitations of this remediation technology and explores the efficacy of intrinsic bioremediation in achieving prescribed remediation endpoints.

174 Enhancing Intrinsic Anaerobic BTEX Biodegradation. Reinhard, M.R.*; Hopkins, G.D. Stanford University, Stanford, CA; LeBron, C.A. NFESC, Port Hueneme, CA. Anaerobic processes play a significant role in the intrinsic bioremediation of gasoline contaminated groundwater. In some instances, it may be necessary to enhance intrinsic biodegradation rates, for instance in cases where the groundwater flow is too small to naturally supply electron acceptors and remove inhibitory byproducts. Field studies were conducted at the Seal Beach Naval Weapons Station in Southern California to demonstrate that intrinsic rates can be enhanced by amending electron acceptors nitrate and sulfate. Three slugs of groundwater augmented nitrate and/or sulfate as the electron acceptors and bromide as a conservative tracer were injected into three test zones located within an anaerobic plume of a gasoline spill. Following injection, groundwater samples were withdrawn from multi-level observation wells and analyzed for BTEX compounds and electron acceptors. Composite and depth-specific samples were analyzed for carbon dioxide, methane, BTEX, nitrate, sulfate and bromide. BTEX behavior indicates that pools of BTEX compounds are present that are not flushed out efficiently. Nitrate utilization was rapid and sulfate utilization. Data show that a significant electron acceptor demand has to be met before BTEX removal is stimulated.

175 Bioaccumulation and Biotransformation of Pyrene by the Freshwater Oligochaete *Limnodrilus hoffmeisteri*. Millward, R.N.*, Fleeger, J.W., Reible, D.D., Keteles, K.A. Lousiana State University, Baton Rouge, LA. Oligochaetes are often found in very high densities in organically-enriched aquatic sediments. Many species bioturbate these habitats by bulk ingestion, which are then defecated at the surface. During this process the common oligochaete *Limnodrilus hoffmeisteri* significantly reduces the sediment concentration of pyrene in spiked sediments as a result of burrowing as well as metabolization. These results suggest that oligochaetes may play a significant role in the transport and biodegredation of PAH-contaminated sediments in the field, and might show some potential in passive bioremediation measures. Experiments with a concentration series of ^{14}C-labelled pyrene were designed to evaluate the fate of pyrene during this feeding process, calculating the biota-sediment accumulation factor (BSAF) to establish bioaccumulation rates and analyzing the produced fecal matter to estimate the net reduction in pyrene content after passage through the gut. Groups of *L. hoffmeisteri* were placed into tubes containing 0, 25, 100, 250, 690 or 2000nmol/gdw of radiolabelled and non-radiolabelled pyrene, and allowed to burrow. Feces and animals were collected from tubes after 2, 5 and 10 days; feces were analyzed for pyrene and organic C content, and animals for pyrene, lipid content and wet weight. Results indicate rapid bioaccumulation of pyrene, particularly at high sediment pyrene contents. The fecal pyrene content in the two highest treatments peaked after 5 days before showing a marked decrease by day 10. This decrease in fecal pyrene content might be a result of increased metabolism with time, or indicate bacterial breakdown of ^{14}C-labelled pyrene into more water-soluble breakdown products. Additional experiments with radiolabelled pyrene are planned to study the biotransformation of the congener into metabolic breakdown products.

176 Biostabilization for Remediation of Pcb and Pah Contaminated Soils and Sludges. Smith, J. R. *, Tabe, M. E., Aluminum Company of America, Alcoa Center, PA. Ongoing bioremediation research is supporting the concept of using engineered land treatment to provide for both the reduction and stabilization of organics in soil/sludge materials to achieve environmentally acceptable end-points (EAEs); thus the term *biostabilization.* Here, biostabilization is defined as a two-step process. Step one entails *active* land treatment for a 2 to 4 month period to achieve reduction of the more bioavailable and leachable (i.e., labile) organics. Subsequent step two involves *passive* (i.e., intrinsic) bioremediation of the remaining organics as they slowly become bioavailable over time. This passive step accounts for continued organic reduction over a period of years and also provides a mechanism to help insure that remaining organics are biodegraded before moving any measurable distance in the subsurface. From a full-scale remediation perspective, certain engineering controls and on-going monitoring may be required during the passive remediation time frame to prevent direct contact of the material from a risk management perspective. However, these controls can be removed as the material achieves certain organic reductions with time. The Aluminum Company of America (ALCOA) is actively involved in two pilot programs that are generating data which supports the concept of biostabilization of PCBs and/or PAHs in soil/sludge materials via engineered land treatment. For total PAHs, active land treatment followed by 3 to 10 years of passive bioremediation achieved between 90 to 96 percent reductions in soil concentrations and greater than 99 percent in water leachable concentrations. Here, total PAH untreated soil concentrations ranged between 1,500 to 3,000 mg/Kg-dry weight and untreated soil water leach concentrations ranged between 0.1 to 1 mg/L. For total PCBs, soil concentrations were reduced between 32 to 48 percent and water leachable concentrations were reduced between 71 to 76 percent. Here, total PCB untreated soil concentrations ranged between 8 to 12 mg/Kg-dry weight and untreated soil water leach concentrations averaged approximately 0.001 mg/L. In addition, treated soil showed no eco-toxicity based on earth worm mortality, root elongation and Microtox testing with untreated soil exhibiting complete toxicity. Lastly, the order of reductions for individual PAHs and PCB compounds agrees with that expected from thermodynamic theory and sorption/biodegradation modeling. The presentation will discuss the basic concepts of biostabilization for PCBs and PAHs and summarize the pilot results to date in the concept of sorption and biodegradation principles.

177 Effect of Land-Biotreatment on PCB Leachability and Risk from Field Contaminated Sediments. Upal Ghosh, Carnegie Mellon University, A. Scott Weber and James N. Jensen, State University of New York at Buffalo, and John R. Smith, ALCOA. Sediments obtained from storage lagoons containing significant amounts of hydraulic oils were subjected to bioremediation in pilot-scale land biotreatment units to assess how biodegradation affects PCB desorption and leachability. During operation of the pilot scale land bioremediation unit, reductions in PCB sediment concentrations of 29% and 46% were achieved for 3 months active biotreatment and after 21 months of subsequent passive biotreatment, respectively. PCB desorption characteristics of collected field samples were studied under batch and continuous flow leaching column conditions. During batch desorption testing, di and trichlorobiphenyl aqueous concentrations decreased with biotreatment as expected, but tetra, penta and hexachlorobiphenyls concentrations were found to increase. Similar trends were observed during operation of the continuous flow columns. Reductions in leachate concentration of lower chlorinated PCBs with a concurrent increase in higher chlorinated congeners as a function of biodegradation was related to the changes in hydraulic oil PCB mole fraction. Loss of lower chlorinated PCBs through bioremediation and higher leaching rates, increased the mole fraction and thus aqueous solubility of higher chlorinated PCBs Based on toxicity factors for each of the PCB homolog groups, leachate toxicity was calculated to increase with biodegradation. Accordingly, study results suggest that biodegradation as a remediation strategy may result, at least for the short term, in an increase in higher chlorinated PCBs and leachate toxicity even though decreases in total PCB concentration are achieved.

178 Changes in Soil Toxicity and Bioavailability during Bioremediation of Aged Petroleum Hydrocarbon Contaminated Soils. Huesemann, M.H.*, Hausmann, T., Gilfoil, T., and Fortman, T., Pacific Northwest National Laboratory, Marine Science Laboratory, Sequim, WA. A set of different soils or model soil particles (i.e., 2 quartz sands, kaolinite clay, montmorrilonite clay, peat, 2 silica gels, a Richland topsoil, and non-porous glass beads) were spiked with a crude oil (5% wt) and aged for more than 2 years in the laboratory. These aged soils, plus an aged bunker C contaminated soil from a Navy site, were subjected to bioremediation treatment in slurry reactors. The changes in soil toxicity during bioremediation were periodically measured using the solid-phase MictrotoxTM assay. Slurry samples were incubated with semi-permeable membrane devices (SPMDs) to determine the fraction of hydrocarbons that may be bioavailable to earth-dwelling species. Slurry samples were also analyzed for parent and alkylated PAHs and biomarkers (e.g., hopane) to determine the bioremediation kinetics of selected hydrocarbons. Abiotic release rates of PAHs from the soil slurries were measured using XAD-2 resin as a sorbent. Preliminary results indicate that in almost all slurries, soil toxicity decreases significantly during the first few months of bioremediation and appears to approach the toxicity level of clean background soils. As expected, high molecular weight PAHs biodegraded more slowly than low-molecular weight ones. The biodegradation and abiotic release rates for specific PAHs will be compared to determine whether the bioremediation process is reaction-rate or mass-transfer limited (i.e., limited by PAH bioavailability to microorganism). Finally, data from the SPMD exposure experiments will be used to evaluate how hydrocarbon bioavailability is affected by differences in soil type.

179 Organic Phase Resistance to Dissolution of Polycyclic Aromatic Hydrocarbon Compounds, Ortiz, E., Kraatz, M., Luthy, R. G.*, Carnegie Mellon University, Pittsburgh, PA 15213 The dissolution of naphthalene, phenanthrene, and pyrene from viscous organic phases into water was studied in continuous-flow systems for time periods ranging from several months to more than one year. By selecting nonaqueous phases from low viscosity to semisolid, i.e., from a light lubricating oil to paraffin, the governance of mass transfer was shown to vary from water-phase control to nonaqueous-phase control. An advancing depleted-zone model is proposed to explain the dissolution of PAHs from a viscous organic phase wherein the formation of a depleted zone within the organic phase increases the organic-phase resistance to the dissolution of PAHs. The experimental data suggest the formation of a depleted zone within the organic phase for systems comprising a high viscosity oil (~1000 cp at 40C), petrolatum (petroleum jelly), and paraffin. Organic-phase resistance to naphthalene dissolution became dominant over aqueous-phase resistance after flushing for several days. Such effects were not evident for low viscosity lubricating oil (86 cp at 40C). The transition between aqueous-phase dissolution control and nonaqueous-phase dissolution control appears predictable, and this provides a more rational framework to assess long-term release of HOCs from viscous nonaqueous phase liquids and semisolids.

180 Application of Soil Biokinetics and Bioavailability Testing Protocol to Enhance Bioremediation and to Achieve Environmentally Acceptable Endpoints in PAH Contaminated Aged Soils. Tabak, H.H., U.S.EPA, Cincinnati, OH; Govind, R., Song Q., and Guo, J., University of Cincinnati, Cincinnati, OH. Bioremediation of polluted soil requires a fundamental systematic multi-level protocol, based on four types of bioreactor systems, was developed to determine the biokinetic parameters of the suspended and immobilized indigenous microbiota, the transport and diffusivity parameters of soil polycyclic aromatic hydrocarbon (PAH) contaminants and oxygen limitation in freshly PAH spiked and PAH contaminated aged soil matrices. The four soil bioreactors used were soil slurry, wafer, porous tube and column reactors. Bench-scale, soil slurry, wafer, porous tube and column bioreactor systems, which mimic field-scale *ex-situ* bioslurry, land farming and *in-situ* bioremediation, respectively, are used in conjunction with respirometry, to quantitatively assess biodegradation rates in dispersed, compacted and intact soils, as determined by oxygen uptake and continuous, automated and simultaneous carbon dioxide evolution measurements. Abiotic adsorption and desorption experiments were conducted to obtain nonlinear isotherms described by the Freundlich isotherm equation. Detailed mathematical models were developed for the four bioreactors and abiotic adsorption and desorption studies together with cumulative oxygen uptake and carbon dioxide evolution data are used to derive the PAH contaminant and oxygen diffusivities in the freshly PAH spiked and PAH contaminated aged soil matrices. The biokinetic parameters obtained, are the Monod kinetic constants for PAH contaminant biodegradation in the soil and aqueous phases. Studies included the use of microcosms to qualitatively assess the biodegradation rates and testing with radiolabeled compounds to determine mineralization. Studies with PAH contaminated Reilly Tar soils and Calhoun Park soils were undertaken to determine the effect of soil properties on the sequestering process that leads to diminished bioavailability; to evaluate the effect of chemical inducers, surfactants, nutrients, cometabolites, inoculum amendments and moisture content on the rate and extent of PAH biodegradation; and to establish the best attainable environmental endpoints for PAH pollutants in aged soils. Modeling procedures, applied to the four experimental schemes, proved useful strategies for determining the biokinetic, transport and diffusivity parameters for PAHs in freshly spiked and aged soils. Values of diffusion and mass transfer coefficients for the PAHs studied, were optimized by the adaptive random search method. The intrinsic transport and biokinetic parameters were derived from experimental data using nonlinear regression analysis and can be used to model the kinetics of on-site and *in-situ* bioremediation.

181 Pentachlorophenol as a Case Study in the Evaluation of Chemicals as Persistent Organic Pollutants (POPs). Woltering, D.M.*, Chrostowski, P.C., The Weinberg Group, Washington, DC. A number of chemicals, mostly chlorinated hydrocarbons, were evaluated under the recently completed United Nations Economic Commission for Europe (UNECE) Protocol on Persistent Organic Pollutants (POPs). The scientific determination of whether or not a chemical qualified as a POP was based on factors including: persistence, bioaccumulation, ecological and/or human toxicity, and its potential for long-range trans-boundary atmospheric transport. Each of the factors has associated guidance values that are considered during the evaluation process; for example, a half-life in sediment of 6 months or a log $K_{OW} \geq 5$. Risk profiles were prepared for each chemical taking all of the available factors into account. This presentation will cover the basics of the UNECE POPs evaluation process along with an overview of the candidate chemicals and a case study detailing the evaluation of pentachlorophenol. Pentachlorophenol was initially among the POP candidate chemicals but it does not have the environmental behavior characteristics of the other chemicals that were ultimately included on the UNECE POPs list. Pentachlorophenol has relatively low persistence and bioaccumulation potential and there is no evidence of long-range trans-boundary atmospheric transport.

182 Case Study on the Application of the Adult Lead Model. Shepherd, D.C.*, Srinivasan, K.*. Woodward-Clyde International-Americas conducted a site-specific human health risk-based assessment for a port facility on the Gulf Coast of Mississippi. The compound of concern was inorganic lead in soils. Lead contamination in soils has historically been evaluated using EPA's Integrated Biokinetic Uptake Model (IUBK), which is focused on lead contamination in residential settings where children are the most sensitive potential receptor, due to physiological differences between children and adults. In order to more effectively address the potential for lead to adversely affect an adult population, the Adult Lead Model was developed by EPA. As of the time this project was conducted, the Adult Lead Model had not been utilized outside of the EPA CERCLA/Superfund program. The Adult Lead Model was applied to the port facility site for the protection of adult workers at the facility for potential exposure to lead in soil. Site-specific inputs were used in the Adult Lead Model. The calculated site-specific risk-based cleanup level for the port facility was 9648 mg/kg (ppm) in soil. In subsequent negotiations with the state agency (MOPC), the cleanup level for the port facility was identified to be 2000 mg/kg (ppm) for onsite soil. This is an increase from the originally identified cleanup level of 400 ppm, based on MOPC's policy at the time of negotiations. This increase resulted in a savings of approximately $470,000 in potential remediation costs. Woodward-Clyde also successfully negotiated a voluntary consent agreement between MOPC and the port which assured MOPC that the owner (the port) would cleanup the facility to residential levels should the facility change use, ownership or control for any reason in the future. Thereby, the current industrial use of the site was considered enforceable and was the basis for approval of the current remedial action for lead in soils.

183 The Evolution of Bio-engineering Strategies at the Year 2000 Olympic Site. Short, K., Olympic Co-ordination Authority, Sydney. When Sydney submitted its bid to host the Year 2000 Olympic Games, *Environmental Guidelines* were included. Preparation for the event included a $A137million remediation program which is now near to completion. Clean-up was essential as the 760 hectare site is a former dump, containing 9 million cubic metres of domestic, commercial and industrial wastes. Originally, the area featured wetlands, waterways, saltmarsh, open grasslands and mixed hardwood forest but land reclamation and waste dumping let to environmental degradation, including pollution with hazardous wastes. Clean-up began several years before the Olympic bid and even at this early stage, engineering works proceeded in tandem with investigations of tidal and ground-water characteristics, habitat and biota. In addition, community workshops identified water quality as the top priority. A risk reduction strategy was chosen as most suitable for the Olympic clean-up. It was based on the reduction of toxicant bio-availability through the elimination or closure of those pathways which generated significant potential for human and environmental exposure. No waste was removed from the site, since a commitment to on-site management had been made. This apparent constraint functioned as a positive for the environment since it encouraged engineers to consider the potential role of biological processes in the delivery of a safe site. In March, 1998, a $A11.6million enhanced remediation strategy was announced. It included a three-tiered monitoring program and community validation program featuring GIS technology. Clean-up has thus evolved from a "best practice" engineering "cap and contain" program to a progressive bio-engineering process that includes biological treatment of air, soil and water. The refinement of this program has the capacity to provide long-term benefits for the environmental integrity and human health.

185 An Integrated Risk Assessment to Investigate Pollutant Exposure and Effect Due to a Coastal Industrial Development in Thailand. Srinetr, V.*, Chongprasith, P., Pollution Control Department, Bangkok, Thailand. An ecological risk assessment is developed to clarify and resolve unsettled issues on metals and petroleum hydrocarbons contamination at Map Ta Phut Industrial Coastal Development, Eastern Seaboard, Thailand. The previous studies (1995-1997) cannot clarify the extent of contamination due to a high variation and limitation of data. This study (in 1998) applies the concept of ecological risk assessment. The study aims to integrate several approaches ; sediment chemistry, seawater chemistry, macrobenthic community and biomonitoring. The extensive sampling design is carefully set up to obtain statistically reliable data for interpretation. The study design is based on a revised conceptual model of the study area. The contaminant pathways are hypothesized. The environmental compartments; including seawater, surface sediment and aquatic species, are synoptically monitored for exposure assessment. Contaminant release, transport, accumulation and fate are identified by sediment core sectioning. Exposure-Dose-Response Triad (Salarzar and Salarzar, 1996) is exemplified in order to characterize both exposure and effect. The study demonstrates that an integration of synoptic data brings about a better summary of the extent of contamination in the study area. A principle of ecological risk assessment supports to draw up a clearer conceptual model. However, ecological risk assessment is complicated, but it can be simplified whenever measurement endpoints are appropriately selected. The risk characterization, the final phase, still includes a high uncertainty. The benchmark concentrations of pollutants are not yet nationwide accepted. Risk estimate need a further study. This study becomes a first example, which ecological risk assessment is applied in Thailand.

186 Re-evaluation of Site Data Following a Screening Level Risk Assessment. Roy, M.M.* and Shaw, T.A., Radian International, Austin, TX; Rak, A., Air Force Center for Environmental Excellence, Brooks Air Force Base, TX. A screening level ecological risk assessment was conducted using surface water, sediment, and surface soil constituent concentrations for ecological habitats. Constituents detected were metals, organochlorine pesticides, and polycyclic aromatic hydrocarbons. If ecological risk was determined at a habitat, then contamination sources were further investigated. To better understand the possibility of false positive risk indicated by background concentrations of metals, data were evaluated on a facility wide basis to determine if habitats contained elevated concentrations of metals. Data were plotted using box plots and a quantile plot. Quantile plots were prepared for log-transformed and untransformed concentrations. Hazard quotients from the ecological risk assessment were reviewed in conjunction with the plots. For some metals, such as aluminum, the plots showed that background concentrations fell within the range of habitat area concentrations indicating a possible false assessment of risk for the habitat when compared to the background area. For other metals, such as arsenic, there are distinct groupings of most habitat areas and background concentrations indicating elevated concentrations at some habitat areas. Those locations with elevated arsenic concentrations also had hazard quotients greater than one. The plots were also used to identify concentrations considered statistical outliers. The combination of the hazard quotients and the data evaluation indicates that, for arsenic, the risk estimate appears to be valid. Additionally, influences not accounted for in the model were explored; these included the total organic carbon content of the soil and the form of the metal used in the benchmark toxicity study. The views in this abstract/presentation do not necessarily represent the views of the agency or the United States government.

187 Development of Assessment Endpoints Based on Wildlife Functional Grouping for Use in Ecological Risk Assessment Guidance in Alaska. Brewer, R.A.*, Witter, M.N. Birkner, P.D., Shannon & Wilson, Inc., Seattle, WA, Himmelbauer, L., Alaska Department of Environmental Conservation, Juneau, AK. The problem formulation phase of an ecological risk assessment includes development of assessment endpoints. These endpoints define the ecological values that are to be protected at a given hazardous waste site, and they allow estimated potential ecological impacts to be related to future risk management decisions. A systematic method was developed for defining wildlife functional groups, related assessment endpoints, and indicator species for use in ecological risk assessments. The functional grouping strategy included organization of plant, invertebrate, fish, and wildlife species by type (i.e. mammal, bird, etc.) and foraging trophic level, and then subdividing these groups by foraging habitats and size. Assessment endpoints were then defined for each functional group based on ecological function and societal value. Stakeholders (including industry, Alaska native groups, the military, recreational and commercial associations, environmental protection groups, and state and federal agencies) provided critical input on cultural and societal concerns that needed to be addressed by the assessment endpoints. One or more indicator species were then chosen to represent each assessment endpoint. Cultural and societal concerns were considered in selecting indicator species, along with ecological relevance and the practical limitations posed by current ecological risk assessment methodologies. Flexibility was retained to realistically reflect site-specific conditions. This strategy was used to define default assessment endpoints and indicator species for ten regions in Alaska, but is applicable elsewhere. The selected assessment endpoints are intended for inclusion in ecological risk assessment guidance in Alaska.

188 Application of Patch and Metapopulation Dynamics to the Understanding of Risks at a Landscape Level. Landis, W.G.*, Lenart, L., Macovsky, L.R., Institute of Environmental Toxicology and Chemistry, McLaughlin, J.F., Huxley College, Western Washington University, Bellingham, WA. One of the classical problems of environmental risk assessment is the extrapolation of concentration-response data to risks in patchy environments. Spromberg, Johns, and Landis have modified the metapopulation models of J. Wu and colleagues to incorporate interaction with a toxicant or other stressor. The initial research demonstrated the importance of initial conditions, distance between patches, and configuration of the landscape. Multiple outcomes were possible from the same initial conditions. This preliminary work led to the Action at a Distance Hypothesis. The hypothesis states that impacts upon one patch can affect distant patches not coming into contact with a stressor. Macovsky confirmed this finding using a constructed metapopulation of the flour beetle, *Tribolium castaneum*. Mortality in patch-1 of a linear metapopulation alters the age structure of the population in the patch farthest from the initial stressor. This finding confirms the Action at a Distance Hypothesis for this experimental metapopulation. More recent work by Lenart and Landis expanded the models to examine the alterations in impacts due to different patterns of concentration-response curves, patch configuration, and whether a source or sink patch is dosed. McLaughlin is expanding this work to large-scale systems using a variety of techniques that detect patterns in nonlinear dynamics. The Action at a Distance Hypothesis offers a powerful tool for understanding the potential impacts and risks at a landscape level. Next steps include further experimental verification, confirmation of the effect in the field, and methods for incorporating these dynamics into ecological risk assessment.

189 Comparison and Use of Deterministic and Probabilistic Ecological Risk Estimates to Guide Wetland Remediation at a Superfund Site. P.M. Rury, D.J. Turton, Arthur D. Little, Inc., Cambridge, Massachusetts, S.C. Svirsky, U.S. Environmental Protection Agency, Boston, Massachusetts, and K. Munney, U.S. Fish and Wildlife Service, Concord, New Hampshire. During Remedial Design at a New England Superfund site, extensive polychlorinated biphenyl (PCB) contamination was found in an adjacent forested wetland downstream from the site. Other contaminants of concern (COCs) in wetland sediments are pesticides, chromium, and lead. Grid-based sediment sampling, earthworm COC uptake studies, and assessment of ecological risks served to guide the wetland remedial design. The ecological risk assessment (ERA) integrated deterministic estimates of sediment risk from all COCs to benthic invertebrates, short-tailed shrew, American woodcock, and mink for several different foraging scenarios with probabilistic food chain exposure estimates for the woodcock and mink. There was good agreement between risk estimates from the two techniques. For woodcock, the Monte Carlo exposure analysis indicated a 65 percent probability that the average total site risk, or hazard index (HI), would equal or exceed the average HI for a deterministic scenario where birds foraged in the contaminated wetland for 35 percent of their diet. For mink, the Monte Carlo analysis indicated a 90 percent probability that the average HI would only slightly exceed the deterministic HI when mink obtained their entire diet from the wetland. Comparison of outcomes from both modeling approaches helped evaluate which deterministic foraging scenarios where reasonable and which were unlikely. The resulting estimates and distributions of risk from both approaches were evaluated to develop sediment cleanup goals for all three wildlife species. Given the difficulties of forested wetland remediation, the most realistic exposure models were then used to calculate residual risks from contamination that might remain after focused remediation.

190 A Prospective Risk Assessment of the Potential Ecological Effects from Removal of a Dam on the Rogue River. Sheehan, P.J., Wenning, R.J. and Wright, B.D., McLaren/Hart, Inc., Alameda, CA and Becklin, D.M., Sportfish Heritage, Grants Pass, OR. Over the past year there has been an increasing number of calls from environment advocates for the removal of dams on various rivers in North America, many of which have been in place for more than 50 years. A primary reason stated for such proposals is the desire to achieve a permanent solution to the perceived problem dam impacts on the migration and recruitment success of anadromous fish such as the Coho salmon. Such is the case for the Savage Rapids Dam, an irrigation water diversion on the Rogue River in Oregon. The pressure to remove dams has not, however, stimulated much research on the ecological effects associated with the breaching of these dams. One problem that has yet to be fully evaluated with respect to the proposed removal of these dams, is the potential effects of the release of millions of cubic yards of currently impounded sediment on the ecology and hydrology of these river systems. A prospective risk assessment addressing short- and long-term effects of sediment release is currently being conducted for Savage Rapids Dam and the Rogue River ecosystem. The assessment includes a chemical and physical characterization of impounded sediment, a modeling simulation of the redistribution of sediment in the river with time following removal of the dam, and finally, a characterization of risks to aquatic organism populations associated with potential exposures to chemicals and habitat modifications. Data analysis and modeling will be performed using geographic information system (GIS) tools. Risks will be described in terms of the probability of percentage reductions in fish and macroinvertebrate populations. This risk assessment to be completed this summer, should provide new insights into the types, probability, and magnitude of effects likely to occur as the result of dam removal projects to restore natural river flows.

191 An Overview Of The Effects Of Silver To Fish. Hogstrand, C.*, University of Kentucky, Lexington, KY; Wood, C.M., McMaster University, Hamilton, ON, Canada. The effects of waterborne silver to fish are greatly dependent upon water chemistry. The degree of acute toxicity is dramatically reduced by complexation of the Ag+ ion to organic and inorganic anions. Of these, chloride, sulfides, organosulfides, and DOC are probably the most important in natural environments. Even the site and mode of toxicity may differ depending on if the fish has been exposed to the free Ag+ or to silver in a complexed form. In the freshwater rainbow trout, acute silver toxicity is exclusively caused by the free Ag+ and the gill transport system for Na+ and Cl-, specifically the Na+/K+-ATPase enzyme, is the site of toxicity. While there is evidence that other forms of silver may accumulate in the gill tissue, the effects of these on the target sites are slim or none. In seawater, there is virtually no Ag+ present and acute toxicity likely originates from one or several silver chloride complexes. In marine teleosts, acute silver toxicity arises from inhibition of drinking and chloride uptake across the intestinal wall, which together results in a net loss of water and increased plasma osmolyte concentrations. While the gill is not the primary target for silver toxicity in marine teleosts, it appears to be in the Squaliformes elasmobranch, the spiny dogfish. In both freshwater and marine systems, chronic effects on fish may occur at silver concentrations far below acute values. Waterborne silver seems to be the phase of concern for both acute and chronic toxicity, but it is still unclear whether or not mechanisms for chronic toxicity are the same as those that cause acute toxicity. Supported by the Silver Council, Kodak Canada LTD, and the NSERC IOR program.

192 Silver Toxicity: A Piscine Review. Shaw, J.R.*, Hogstrand, C., Birge, W.J., University of Kentucky, Lexington, KY and Wood, C.M., McMaster University, Hamilton, ON. This presentation will review the acute and chronic toxicity of silver to freshwater and marine fish, including water chemistry parameters affecting toxicity. In freshwater, acute silver toxicity generally is a function of free silver ion concentrations (Ag^+). However, the current U.S. EPA acute criterion is hardness based and does not account for this fact. While hardness offers modest protection against silver toxicity, other parameters (chloride, DOC, sulfide) appear to be far more important modulators of toxicity. Typical 96-h LC50 values (mg/L) for silver in freshwater range from 0.006 (rainbow trout, fathead minnow) to 0.065 (bluegill). Chronic exposures have yielded MATC that range from 0.0001 to 0.005 mg/L and acute to chronic ratios (28-50, rainbow trout; 100, fathead minnow) are high compared with other metals (Cu ~ 3). In seawater Ag^+ is essentially absent and different silver chloride species predominate. Reported 96-h LC50 values (mg/L) for marine fish span three orders of magnitude, 0.008 (summer flounder) to 2.7 (mummichog). The current acute criterion for silver in seawater is 0.0023 mg/L, and does not account for parameters that effect acute silver toxicity in marine fish (salinity, ammonia, thiols). There have been few sub-chronic/chronic studies in seawater. Reductions in hepatic enzyme activities have been observed in winter flounder (aminotransferases) and mummichogs (catalase, xanthine oxidase) following exposures to 0.01 and 0.03 mg/L, respectively. Reductions in hatching success have been observed at concentrations as low as 0.09 mg/L for winter flounder, while no effects on hatching were observed at concentrations as high as 5.0 mg/L for sheepshead minnows. The 28-d LC50 for early life stages of the sheepshead minnow was 1.09 mg/L. Currently, there is no chronic criterion for silver in seawater.

193 Effects of Water Characteristics, Sediments, and Chemical Form on Chronic Silver Toxicity. Davies, P.H.*, Brinkman, S.F., and McIntyre, M.W., Colorado Division of Wildlife, Fort Collins, CO. Research was performed to evaluate effects of water characteristics, sediments, and chemical form on the chronic toxicity of silver. We also determined if EPA's acute silver criteria would protect aquatic life chronically exposed to silver. Early life stage (ELS) toxicity tests using silver nitrate were conducted on brown and rainbow trout. Chronic values derived for both species were virtually identical. These were 0.20, 0.50, and 1.08 μg Ag/liter at hardness/chloride concentrations of 25/3, 200/13, and 450/25 mg/liter, respectively. Toxicity experiments were also conducted with water quality and streambed characteristics typical of erosional, high mountain and depositional, plains sediments. Using silver nitrate, a flow-through diluter delivered 0.6 μg Ag/liter in a water hardness of 50 mg/liter to three large equilibrium tanks containing mountain stream sediments and control tank s. Water retention time in the six sediment tanks was 33 hrs, prior to rainbow trout exposure. Similar experiments with rainbow and brook trout were conducted with plains sediments at a water hardness of 450 mg/liter hardness and 27 μg Ag/liter, equal to EPA's criterion. There was little complexation or adsorption of silver in the sediment tanks. With mountain sediments, we obtained 56% mortality in 13 days exposure to 0.6 μg Ag/liter. At 27 μg Ag/liter, in plains sediment experiments, 100% mortality occurred within 15 and 20 days with brook and rainbow trout, respectively. In similar experiments with plains sediment, using silver sulfate, a considerably less soluble form of silver, we found silver sulfate to be significantly more toxic than silver nitrate with 100% rainbow mortality within four days. In an experiment with silver iodide, one of the most insoluble forms of silver, we obtained 18 % chronic rainbow trout mortality at 0.08 μg/liter.

194 The Physiological Costs of Silver Acclimation to Rainbow Trout. Galvez, F.*; Wood, C.M., McMaster University, Hamilton, Ontario. The physiological effects of chronic exposure to $AgNO_3$ in moderately hard fresh water (~120 mg/L as $CaCO_3$) were investigated in juvenile rainbow trout (*Oncorhynchus mykiss*). Two separate 28-day exposures were performed at silver concentrations of 0.5 and 2.0 μg/L under flow-through conditions. Exposure to 0.5 μg/L Ag resulted in a 14.9% increase in food consumption, whereas growth rates (weight) remained unaltered. Both plasma Na^+ and Cl^- levels were decreased by approximately 10% by day 16 of the exposure. In comparison, chronic exposure to 2.0 μg/L Ag resulted in a 28.8% decrease in food consumption and a 43.0% reduction in growth rate. Plasma [Na^+] was decreased by 18.3% and plasma [Cl^-] was reduced by 12.2% at day 7. At both silver concentrations, plasma ion concentrations returned to control levels by the end of the experiment suggesting acclimation to silver may have occurred. Exposure of rainbow trout to these low levels of Ag elicited an ionoregulatory disturbance qualitatively similar but quantitatively smaller, than that seen in other studies at acutely lethal silver concentrations. Present studies are being conducted to assess the physiological costs of silver acclimation in rainbow trout and determine the Ag levels in water required to invoke acclimation. Initially, fish are being exposed to 0.1 and 1.0 μg/L Ag for 20 days; the lower Ag level was proposed, but later withdrawn, by the US EPA as the chronic criteria in fresh water. Over the 20-days, acute toxicity tests are being performed to determine whether the fish are becoming more resistant to acutely lethal effects of the metal. The degree of acclimation is being further tested by quantifying branchial Na^+ influx, branchial Na^+/K^+ ATPase activity and plasma Na^+ and Cl^- levels. (Supported by Kodak Canada and the NSERC IOR Program)

195 Influence of Exposure Route on Sublethal Toxicity of Silver and Selenium to Zooplankton. S.E. Hook and N.S. Fisher. Marine Sciences Research Center, State University of New York, Stony Brook, NY. Laboratory based toxicity tests must use environmentally realistic concentrations, consider all routes of exposure the organism is likely to encounter, and monitor ecologically relevant end points if they are to accurately assess contaminant effects. Previous work done in our laboratory has shown that silver exerts a toxic effect on zooplankton by depressing egg production at environmentally realistic concentrations. These effects occurred at 1 nM for marine copepods and at 500 pM for freshwater daphnids only when the animals were exposed to the Ag in food. Dissolved phase exposure had no discernible effect on toxicity. Similar tests were conducted to determine if selenium exerts comparable effects in zooplankton. In one set of experiments, marine copepods and freshwater cladocerans were exposed to solutions of 0, 0.5, 1,2,5 and 10 nM Se as selenite. In another set of experiments, animals were fed phytoplankton cells grown in these same concentrations. To assess toxicity, egg production, hatching rate and growth rate of juveniles were measured. These results will be compared to the findings for Ag. To determine the mechanism of sublethal toxicity, protein analysis of eggs produced by animals exposed to either silver or selenium as well as control animals were examined via gel electrophoresis.

196 Mechanisms of Ionic Silver, Ag^+, Uptake Across the Gills of Freshwater Rainbow Trout. Bury, N.R.*, McGeer, J.C., and Wood, C.M., INRA, Rennes, France and McMaster University, Hamilton, Canada. Ionic silver, Ag,$^+$ concentrations have been shown to correlate with silver toxicity in rainbow trout. The purpose of this study was to investigate the mechanism(s) by which Ag^+ enters the fish. Apical uptake was studied *in vivo* using adult rainbow trout adapted to softwater, whilst gill basolateral membrane transport was studied using an *in vitro* basolateral membrane preparation (BLMV). Of the three salts, $Ca(NO_3)_2$, KNO_3 and $NaNO_3$ added to the water, only $NaNO_3$ reduced the degree by which the fish accumulated silver. Kinetic analysis shows that the interaction between Na^+ and Ag^+ uptake is competitive. However, at very high Na^+ concentrations (13 mM Na^+) the interaction is both competitive and non-competitive. Phenamil, a specific Na^+ channel blocker, reduced Ag^+ uptake. Taken together these results suggest that apical uptake of Ag^+ is via a Na^+ channel. BLMV studies show that Ag^+ is extruded across the basolateral membrane by an ATP-dependent transport process. This transporter requires the presence of K^+ to function, suggesting that Ag^+ is transported by the Na^+/K^+ -ATPase. (Supported by Kodak Canada Ltd and the NSERC IOR Program)

197 Uptake, Internal Redistribution, and Depuration of Silver in Freshwater Rainbow Trout and European Eel: The Influence of Silver Speciation. Wood, C.M*, McMaster University, Hamilton, ON, Canada; Hogstrand, C. , University of Kentucky, Lexington, KY; Grosell, M. & Hansen, H., Riso National Lab, Roskilde, Denmark. Water chloride protects against the acute toxicity of $AgNO_3$ in freshwater trout by converting the toxic moiety Ag^+ to $AgCl_{aq}$. We therefore examined whether [Cl^-] affects the kinetics of silver in two species differing 10-fold in their sensitivity to Ag^+, *Oncorhynchus mykiss* (sensitive) and *Anguilla anguilla* (insensitive). Fish were exposed for 24 h to 1.8 :g L^{-1} total Ag, added as $AgNO_3$ and labelled with ^{110m}Ag in synthetic soft water of 2 Cl^- levels (50 or 1000 :mol L^{-1}). At low [Cl^-], Ag was present mainly as Ag^+, and at high [Cl^-], mainly as $AgCl_{aq}$. Eels internalized less Ag than trout, despite similar gill loading. Gill Ag accumulation was triphasic with small increases to 6 h, larger increases at 12 h, and lower levels at 24 h, indicating the start of depuration despite continued exposure. $AgCl_{aq}$ loaded into the gills to a greater extent than Ag^+. $AgCl_{aq}$ depurated faster than Ag^+ from trout gills, whereas Ag^+ depurated faster from eel gills. Most (>80%) of the Ag loaded into gills over 24 h was cleared rapidly (5-10 days) in both species. Internal redistribution patterns were largely independent of speciation. The liver appeared to be the final sink for internalized Ag, with hepatic Ag levels increasing or stable over the entire 67 day post-exposure period, whereas Ag levels in other compartments declined. We conclude that gill total Ag levels are not a useful indicator of either the exposure history or the toxic effect of silver in fish. (Supported by Kodak Canada, Kodak U.S.A, AUCC, NSERC IOR, and Riso).

198 Modelling Silver Binding to Gills of Rainbow Trout, Revisited. Rose-Janes, N., and Playle, R.C.* Wilfrid Laurier University, Waterloo, Ontario, N2L 3C5. In November, 1995, *Environ. Toxicol. Chem.* published a paper by Nancy Janes and Richard Playle entitled "Modeling silver binding to gills of rainbow trout (*Oncorhynchus mykiss*); *ET&C* 14: 1847-1858. The model was based on conditional equilibrium stability constants determined for Ag binding to trout gills (log $K_{Ag\text{-}gill}$=10.0), Ag binding to dissolved organic carbon (log $K_{Ag\text{-}DOC}$=9.0), plus Na, Ca, and H^+ binding at the Ag binding sites (log $K_{Na\text{-}gill}$=4.7, log $K_{Ca\text{-}gill}$=3.3, log $K_{H\text{-}gill}$=5.9). Evidence was accumulating that Ag caused disruption in ionoregulation in trout, but few details were available in 1995. Today we know the precise physiological processes affected by Ag at gills of freshwater fish, mainly the inhibition of basolateral Na^+, K^+ ATPase activity. In fish exposed to Ag in softwater we have found that any process which reduces the amount of Ag^+ available to bind to trout gills reduces the ionoregulatory and respiratory effects of Ag. For example, in an exposure to 0.2 μM Ag (as $AgNO_3$), 5.0 μM thiosulphate (log $K_{Ag\text{-}S2O3}$=8.8) kept most Ag off trout gills and out of the plasma, as did 8 mg C·L^{-1} DOC (1.2 μmol·L^{-1} binding sites for Ag). Trout exposed to Ag at 4.7°C accumulated about a third as much Ag on their gills and in plasma as did fish exposed to Ag at 18°C. This reduction occurred because less Ag passed over the gills of the cold fish which had much lower metabolic demands for oxygen, and because the active uptake of ions plus Ag at the gills was reduced (e.g. Q_{10}= 2 to 4 for Na^+, K^+ ATPase). The gill modelling approach does not yet include temperature effects, but does indicate that the order of importance of natural protective agents in water is DOC, Cl, Na, Ca, and H^+, based on their conditional stability constants (log K_{AgCl}=3.3, log K_{AgCl2}=5.3).

199 Application of a Model of Acute Toxicity of Silver to Fish. Paquin*, P., HydroQual Inc., Mahwah, NJ; Wu, B., HydroQual Incorporated, Mahwah, NJ; Santore, R., HydroQual Incorporated, Mahwah, NJ; Di Toro, D. HydroQual Incorporated, Mahwah, NJ and Manhattan College, Bronx, NY. When silver is discharged to a water body, speciation and complexation reactions control its distribution among various organic and inorganic complexes and as ionic silver. It is the distribution of the silver among these forms, as well as the other water quality characteristics of the system, that control its bioavailability. The bioavailability can be evaluated in the context of a biotic ligand modeling framework. In this mechanistically based framework, the biotic ligand, the organism tissue at the site of action, is represented in the same way as any other ligand in solution. It has a characteristic binding site density and conditional stability constants for each of the species with which it reacts. The model simultaneously accounts for the speciation and complexation of dissolved silver and competitive binding of silver and other cations at the biotic ligand. The organism LC50 corresponds to the point where silver accumulation at the biotic ligand reaches a critical level. The silver:gill incarnation of the biotic ligand model has been applied to several datasets for fish. The model will first be used to predict fish gill silver levels over ranges of hardness, DOC, chloride, pH, and alkalinity. It will be shown that the model can also be used to explain the variation in acute toxicity observed in laboratory bioassays with fish, where test conditions including hardness, DOC and chloride were systematically varied. This model has a number of potential applications including use in prediction of water effect ratios (WERs) from site water chemistry, and more generally, for ecological risk assessment purposes.

200 Fate and Effects of Silver in Aquatic Environments: An Overview. Di Toro*, D. HydroQual Incorporated, Mahwah, NJ and Manhattan College, Bronx, NY; Santore, R., HydroQual Incorporated, Mahwah, NJ; Wu, B., HydroQual Incorporated, Mahwah, NJ; Paquin, P., HydroQual Inc., Mahwah, NJ. This presentation summarizes the results of a focused, multi-disciplinary research effort. The research was directed at gaining an improved understanding of the fate of silver in aquatic environments, how it reacts and is transported within a water body, and the manner in which it exerts toxicity. It is well known that when silver is discharged to a water body, speciation and complexation reactions result in the silver being present in a variety of particulate and dissolved forms, and these forms are associated with varying degrees of bioavailability. These processes have been incorporated within a biotic ligand modeling framework that is used to evaluate bioavailability and toxicity. This model simultaneously accounts for speciation and complexation of dissolved silver in the water column and competitive binding of silver and other cations to a biotic ligand, such as the gill of a fish. Besides hardness, it considers the effects of DOC, chloride, pH and alkalinity on bioavailability and toxicity. The fraction of the silver that sorbs to particulate matter may settle from the water column to the sediment where its fate and effects have also been studied. Typically, in anoxic sediments, the silver reacts with acid volatile sulfide (AVS) to form silver sulfide, an insoluble form that is essentially non-bioavailable and hence not toxic to sediment dwelling organisms. The silver sulfide which is in the aerobic surficial layer could potentially oxidize and be released back to the water column, but recent experimental work has shown that the rate of oxidation is very slow.

201 Evaluation of Contaminated Estuarine Sites Using Sediment Quality Guidelines and Ecotoxicological Assessment Methodologies. Fulton, M., Daugomah, J., Bearden, D., Scott, G., NOS, Charleston, SC. Toxic contaminants may enter estuarine ecosystems through a variety of pathways including nonpoint source runoff, industrial point sources and atmospheric deposition. Contaminants may then become incorporated into estuarine sediments. When sediment contaminant levels become sufficiently high, they may impact resident biota. A variety of approaches have been developed for evaluating sediment quality. These include ERL/ERMs, TEL/PELs and AETs. Other studies have assessed the utility of macroinvertebrate indicators for the evaluation of pollution impacts in aquatic ecosystems. The goal of this study was to evaluate the impact of sediment-associated contaminants in tidal creek systems near Charleston, SC by monitoring the population densities of the grass shrimp, *Palaemonetes pugio,* and to evaluate the results of this monitoring effort with respect to ERL/ERM values determined for sediment-associated contaminants at each of the sites. Three impacted sites and one reference site were selected for study. Grass shrimp populations were sampled monthly using a push-netting approach. Nine 25-m creek reaches were sampled at each site and used to calculate site-specific mean annual densities. Three replicate sediment samples were collected at each site and analyzed for metals, polycyclic aromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs) and pesticides. The data obtained from these analyses were then used to calculate cumulative ERM quotients. There was good agreement between the effects observed on grass shrimp populations at each of the sites and the site-specific ERM quotients. Grass shrimp population densities at the sites decreased as the ERM quotients increased. These findings support the use of both sediment quality guidelines (ERL/ERMs) and site-specific population monitoring to evaluate the impacts of sediment-associated contaminants in estuarine systems.

202 Regional Monitoring for Sediment Contamination in the Southern California Bight. Schiff, K.*, Southern California Coastal Water Research Project, Westminster, CA; and Gossett, R., CRG Laboratories, Inc., Terminal Island, CA. Regional monitoring in the southern California Bight (SCB), accomplished through the cooperative and integrated efforts of 12 diverse agencies, was conducted in the summer of 1994. These large-scale assessments of environmental condition were conducted to enhance typically local, site-specific monitoring. As part of this study, 248 sites were sampled using a stratified-random design and sediments were measured for grain size, TOC/TN, 14 metals, and 52 separate organic compounds including PAH's, DDT's, and PCB's. The results showed that certain geographic regions such as Santa Monica Bay were heavily influenced by anthropogenic activities. Except for grain size, concentrations of nearly every chemical constituent was higher inside the Bay than outside the Bay. Also, regions associated with different types of inputs showed distinct differences. For example, sediments near publicly owned treatment works (POTW) wastewater outfalls were elevated in trace metals such as silver, mercury, as well as $\sum$DDT and $\sum$PCB. Sediments near the largest rivers and creeks in the SCB were elevated in trace metals, particularly chromium, copper, lead, and zinc. Chlorinated hydrocarbons were the most widespread anthropogenic contaminant in the SCB. Approximately 82% of the SCB contained detectable quantities of $\sum$DDT; 64 % of the SCB contained detectable $\sum$PCB. Using iron as a conservative element for estimating the naturally occurring fraction of trace metals, at least 50% of the SCB sediments were enriched in at least one of the metals we measured. Although the areal extent of sediment contamination in the SCB appeared large, widespread biological effects were not observed in the other portions of this study.

203 Identifying Sources of Chemical Risk in Urban Waterways from Stormwater, Sewer Overflows and Sewage Treatment Plant Discharges. Bickford, G.P* and Hansen, J.A., Sydney Water Corporation, Sydney, Australia. Ecological and human health risk assessments were completed for sewer overflows, stormwater and sewage treatment plant (STP) discharges in Sydney, Australia to identify priorities for waterways improvements. The relative risks of a range of chemicals (pesticides, herbicides, PAHs, metals, monocyclic aromatic compounds, halogenated aliphatic compounds and other organics) in urban runoff and sewage discharges to freshwater, estuarine and ocean waterways across the Sydney region were analysed. Risks to aquatic organisms were estimated using hazard quotients derived from measured chemical concentrations in waterways, effluent discharges and sediment together with water quality and sediment quality guidelines. Aquatic life risks were validated using biotic surveys and whole effluent bioassays. Risks to human health were assessed for incidental ingestion of water and fish consumption. The results of the studies indicate that stormwater discharges pose greater risks to aquatic biota than sewer overflows because of greater chemical loads and duration of impact following rainfall. Risks to aquatic biota due to sewage treatment plant discharges were related to the nature of the receiving water body rather than treatment level provided. Potential risks identified for chemicals in discharges to rivers and streams are being verified through a TIE program for 17 STPs. Risks to people from incidental ingestion of water during recreation were negligible; risks due to consumption of fish were similar across waterways and discharge types. The risk assessment results have been used to prioritise STP treatment upgrades and sewer overflow abatement works in the Sydney region.

204 Effects of Acidic Ph on Benthic Macroinvertebrate Communities Collected from Metal-polluted and Reference Streams in Colorado. Courtney, L.A.*, Clements, W.H. Colorado State University, Fort Collins, Colorado. We examined the effects of a novel stressor, acidic pH, on benthic invertebrate communities from streams with different histories of metals pollution: two metal polluted sites on the Arkansas River (AR1 and AR5) and a reference site on the Cache la Poudre River, Colorado. Long-term metals exposure has resulted in tolerance within the Arkansas River communities, and there is concern about costs associated with tolerance. Exposure of invertebrate assemblages to metals in stream microcosms significantly decreased survival only in the Cache la Poudre River assemblage; the AR5 and AR1 mayfly assemblages were more tolerant to the metals mixture. We compared the effects of metals exposure on the three communities to the effects of acidic pH, in similar experimental conditions. Experiments showed site specific differences in responses to acidic pH, which were increasingly evident at finer levels of taxonomic resolution. Multivariate analyses revealed different patterns of response among communities and showed that effects of acidic pH on community composition were greater for the metal-tolerant Arkansas River assemblages. Two-way ANOVA supported multivariate analyses and demonstrated significant site, treatment, and interaction effects on mayfly abundance ($p<0.0001$). The number of mayflies was reduced significantly only in the AR5 assemblage. Differences in effects of acidic pH resulted from differences in community composition. The mayfly assemblage at AR5 was dominated by *Baetis* spp., which were eliminated from all treatment streams. In contrast, mayfly species abundant at the other sites were less sensitive to acidic pH. Concentrations of metals are higher at AR5 than AR1, and the elevated metals concentrations may have played a role in structuring a community tolerant to metals but especially sensitive to acidic pH, a novel stressor.

205 Canadian Program on Aquatic Effects Technology Evaluation. Trudel, L.*, Natural Resources Canada, Ottawa, ON, Canada; McKee, P., Farara, D., and Hart, D., Beak International Inc., Brampton, ON, Canada. Canada is currently reviewing its *Metal Mining Liquid Effluent Regulations* (MMLER) under the *Fisheries Act.* The revised MMLER will include a requirement that mine operators implement environmental effect monitoring of aquatic ecosystems. The Aquatic Effects Technology Evaluation (AETE) Program stem from the cooperation between government and industry to evaluate environmental monitoring techniques to be used by the mining industry and regulatory agencies in assessing the impacts of mine effluents on the aquatic environment. Existing techniques were compared and assessed in terms of benefits/limitations and cost-effectiveness. Recommendations were made on specific methods or groups of methods that will permit accurate characterization of environmental impacts in an overall monitoring strategy. Field surveys and laboratory evaluations of selected methods were conducted from 1995 to 1997 at five mine sites across Canada: three base metal mines and two gold mines discharging in various types of freshwater receiving environment. Monitoring methods included: sublethal toxicity tests of water/effluent using *Lemna minor*, *Selenastrum capricornutum, Ceriodaphnia dubia* and Fathead minnow; sediment toxicity tests using *Tubifex tubifex*, *Hyalella azteca* and *Chironomus riparius*; use of metallothionein as a biomarker and bioaccumulation of metals in fish tissues; use of fish (community structure, growth and organ size) and benthic macro invertebrates (population/community); analysis of dissolved metals in water at ultra low detection limits; application of full and partial metal extraction methods and AVS/SEM ratio in sediments. The results were interpreted to test the relative ability of tools to detect an effect or a relationship between mine exposure and ecological responses. A triad evaluation examined correlations between the chemical, toxicological and biological *arms* to support that metals or contaminants were responsible for biological effects.

206 Use of Monitoring Data in the Calibration and Verification of Environmental Fate Models. Cowan, C.E., The Procter & Gamble Company, Cincinnati, OH, USA; Feijtel, T.C.J., The Procter & Gamble Company, Strombeek Bever, Belgium; Giger, W., Swiss Federal Institute of Water Resources, Dviebendorf, Switzerland; Mathies, M., University of Osnabruck, Osnabruck, Germany; Tanabe, K., National Institute of Environmental Studies, Tsukuba, Japan. One of the key elements in the environmental risk assessment of chemicals is the estimation of the concentration that is likely to occur in the environment, i.e., the Predicted Exposure Concentration (PEC). PECs may be derived from monitoring data or from the predictions of environmental fate models. For new chemicals and for chemicals for which environmental monitoring is not possible due to analytical difficulties and/or cost, environmental fate models are the method of choice. However, concerns have been raised regarding the accuracy of these environmental fate models. For this reason, OECD held a workshop in which guidance was developed on how to 1) chose monitoring data for use in calibration and verification of environmental fate models and 2) fill any data gaps that exist in the monitoring data including evaluating extrapolation techniques. In addition, the workshop participants discussed criteria for model verification and provided guidance in cases when model predictions have not been verified. Finally, the participants recommended ways that monitoring programs could be improved to make them more useful in model calibration and verification. In this presentation, I will present the results of these discussions and relate these to experiences in model calibration and verification using monitoring data.

207 EPA's National Study of Chemical Residues in Fish. Farris, J. M.*, U.S. EPA, Washington, DC. In 1986, the EPA Office of Water conducted a national screening level investigation to determine the prevalence of selected bioaccumulative pollutants in fish, and to correlate elevated levels of fish tissue contaminants with sources of these pollutants. In the 1986 study, fish were collected from 314 locations thought to be influenced by a variety of point and nonpoint sources. A list of 60 target analytes was developed for the study (including dioxins and furans, PCBs, pesticides/herbicides, mercury, and a number of other organic compounds) and analyses were conducted to determine levels of these contaminants in fish tissue. Bottom feeding and gamefish species were collected at each sample site. Results of the 1986 study indicated that target analytes were present in fish tissue at many of the sites sampled, and some of the contaminants occurred at levels posing potential human health risks. The EPA Office of Water has recently started work on a new three year national study of chemical residues in fish designed to answer a number of questions not addressed in the 1986 study. The proposed study objective is to estimate the national distribution of the mean levels of selected persistent bioaccumulative toxic (PBT) chemical residues in fish tissue in estuarine and inland waters of the continental United States. The new study will provide data on tissue contaminants in fish from a greater number of waterbodies than were sampled in 1986. A probabilistic sampling design will be used in the new study to assess the national extent of the fish tissue contamination in U.S. waters. Evaluation of tissue levels of selected pesticides and other bioaccumulative toxics of concern in agricultural and urban areas are of particular interest. The sampling framework will be reviewed by a workgroup and peer group before inititation of the sampling program in 1999.

208 Comparative Effects of Pesticide Runoff From Tomato Crops Grown Under Two Discrete Cultivating Practices on Estuarine Organisms. Hetzer, P.R.*,Chesapeake Biological Laboratory (CBL), Solomons, MD; Brown, S.S., CBL, Solomons, MD; Baker, J.E., CBL, Solomons, MD; McConnell, L.L., USDA, Beltsville, MD. We evaluated the effects of surface-water runoff and sediment deposited from experimental outdoor agricultural plots on four estuarine organisms: the copepod *Eurytemora affinis,* the amphipod *Leptochierus plumulosus*, the hard clam *Mercenaria mercenaria*, and the diatom *Thalassiosira psuedonana.* Experimental plots consisted of two treatments with four replications per treatment. The first treatments utilized a growing technique used widely throughout the U.S. termed *plasticulture,* in which raised beds are covered with black polyethylene to control soil temperature, humidity, and weed growth. The second treatment was a new technique in which a hairy vetch, *Vicia villosa*, was grown on the plot, and then mulched to obtain the same beneficial effects as the plastic. Tomatoes were grown identically (except for mulching methods) in each plot with equivalent pesticide applications. Runoff water was collected approximately twice a month (May-Aug.) following pesticide applications and a subsequent rainfall event (worst case) that produced a significant surface-runoff. Dissolved concentrations of target analytes (endosulfan I and II, endosulfan-sulfate, fenvalerate, esfenvalerate, chlorothalonil, metribuzin, copper) were quantified in water, and sediment samples collected on site. A subsample was then analyzed immediately prior to the beginning of the bioassays to determine any loss during storage. Standard bioassays were conducted to determine lethal and sublethal effects on the test organisms resulting from pesticide exposure. Additional laboratory studies were conducted to investigate the independent and interactive effects of selected pesticides, and to determine the effects of pulsed-exposures on test organisms.

209 Integrated Monitoring and Assessment of Surface Water as a Tool for Informed Decision Making and Sustainability. Michaelidou C.St., State General Laboratory, Nicosia, Cyprus. Human health and well being are linked with the sustainability of our environment, which in turn is dependent on the preservation of the biological chain. Prognosis and prevention of impacts of chemicals on the ecosystem are essential. These can be only achieved through the integration of chemical and biological approaches, which can compliment each other resulting into a robust system for environmental monitoring and hazard identification and evaluation. The development of an innovative monitoring, Early Warning System for surface waters (developed within an EU Life Program) will be presented. To sustain quality and multifunctionality of surface waters, pollution status and trends as well as possible effects on Human Health and the Ecosystem are evaluated. To bridge the gap between ecotoxicological assessment and decision-making, an attempt was made to integrate and express results from 236 individual parameters (chemical, microbiological and biological) with nine quality indices. These are the Industrial pollution (IPI), the Pesticide pollution (PPI) the Nutrient Pollution (NPI), Organic Pollution (OPI), the Bacteriological Index (BQI), the Toxicity (TOX), the Biodiversity (BDI), the Benthic Saprobity (BSI) and the Irrigation Quality Index (IQI). Each index is reflecting a different type of pollution or effect. All indices are integrated in one "yardstick" (the Amoeba) by which the status of water resources and degree of deviation from targets are clearly described. Priorities for policy measures can also be easily identified. Target values were developed based on available information, limits, expert judgment and socioeconomic factors. This innovative assessment tool can facilitate the use of forecasting models and the integration of research results into the decision making process, in particular for Environmental Impact Assessment studies undertaken for new developments within the catchement zones of the dams.

211 Dosimetry of Phototoxic Polycyclic Aromatic Hydrocarbons. Erickson, R.J.*, Ankley, G.T., Kosian, P.A., Makynen, E.A. and DeFoe, D.L., U.S. Environmental Protection Agency, Duluth, MN. The toxicity of some polycyclic aromatic hydrocarbons (PAHs) can increase by an order of magnitude, or more, in the presence of ultraviolet (UV) radiation. A dosimetry model was developed based on damage accumulating with time proportionately to the product of chemical accumulation and light intensity. The acute phototoxicities of selected PAHs to the oligochaete *Lumbriculus variegatus* were investigated in order to test this model. Test organisms were exposed to multiple concentrations of anthracene, pyrene, and fluoranthene in water for 96 h and then to various intensities of ultraviolet light for 96 h in clean water. Time-to-death was found to adhere well to the model, and, based upon measured accumulations of PAHs, anthracene and pyrene were respectively 2.5 and 1.7 times as potent as fluoranthene. The phototoxicities of mixtures of these chemicals were also evaluated and were predicted well using the model if it was assumed that the damage accruing from the individual PAHs was additive.

212 The Effect of Light Spectra on the Photo-Induced Toxicity of PAHs: Implications for Risk Assessment. Diamond, S.A., Mount, D.R., Burkhard, L.P., Ankley, G.T. U.S. EPA, Duluth, MN. A major factor in PAH photo-activated toxicity is the intensity and spectral distribution of solar radiation reaching potentially affected organisms and tissues. To demonstrate the effect of light spectra on photoactivated toxicity brine shrimp assays were conducted with pyrene, anthracene and fluoranthene under lighting regimes varying either in total UV-A intensity or in spectral distribution. The effect of spectral distribution ranged from none to a very significant 2x variation in toxicity, depending on the PAH tested. Differences in toxicity in assays where intensity was varied and spectral distribution was uniform were strongly related to total UV-A intensity ($R^2 >$ 0.98 in all assays). The results of these two types of assays can be accurately predicted by the multiple of spectral intensity and molar absorptivity of each PAH, integrated over all UV-A wavelengths. However, the fit of such models is limited by knowledge of the light spectra actually present at the site of activity (the PAH-biomacromolecule complex) and the absorbance of PAHs in those complexes. To illustrate the potential effect of spectral variation in contaminated sites, several locations in northern Minnesota were examined. The potential for photoactivation of the three PAHs tested varied dramatically among sites, from clear water locations in which activating wavelengths were only slightly attenuated, to high-DOC sites where little activating radiation was apparent even at moderate depths. These results will be discussed in the framework of dose estimation in risk assessment of PAH photo-activated toxicity. Visual inspection of light spectra present in various aquatic habitats, and within brine shrimp tissues, as well as variation in *in-situ* PAH absorbance spectra will be presented.

213 Modeling Site Specific Phototoxicity of Pah in Natural Waters: Case Study of Lake Tahoe, California/nevada, Usa. J.T. Oris[1], A.C. Hatch[2], J.E. Weinstein [1], R.H. Findlay [1], S.A. Diamond [3], G.A. Burton [2], and B. Allen [4]. [1] Miami University, Oxford, OH 45056; [2] Wright State University, Dayton, OH; [3]National Research Council (U.S. EPA), Duluth, MN; [4] Univ. California-Davis, Davis, CA. A focus of recent research has been the consideration of solar UV radiation and photo-induced toxicity in the development of site specific water and sediment quality criteria of PAHs. Ample evidence exists from laboratory research that indicates reasonable hazard estimates can be obtained from a simple model that accounts for the body residue and photochemical reaction efficiency of a particular suite of PAHs and for the incident level of UV-A radiation. However, no tests of the model have been conducted under ambient field conditions of extremely low PAH (ng/L) and high UV radiation intensities. Experiments were conducted at Lake Tahoe to assess the predicted impact of ambient levels of motorized watercraft emissions on zooplankton and fish larvae. Water was collected daily from a nearshore site and an offshore site, representing areas of high and low boating activity, respectively. Water from the sites was returned to shore and used in standard effluent toxicity tests modified to account for ultraviolet radiation-induced toxicity of PAHs. Separate concentration-response exposures using fluoranthene were also conducted. Results from the tests indicated that ambient levels (10-70 ng/L) of PAHs, putatively derived from motorized watercraft emissions, caused significant phototoxicity to fish and zooplankton. Model predictions of levels of effect in both ambient toxicity testing and concentration-response studies were not signficantly different from test results, indicating that site-specific hazard assessments can be predicted for phototoxic compounds.

214 Direct and Indirect Effects of UV Radiation on Plants and Phytoplankton: PAH Photoinduced Toxicity and UVA/UVB Damage. B.M. Greenberg, C.A. Marwood, T.S. Babu, C.L. Duxbury, X.-D. Huang, A. Mallakin, Dept. of Biol., Univ. of Waterloo, Waterloo, ON, Canada. Because plants live in high photic environments, they are exposed to UV radiation. UV can negatively impact on plants via both exogenous and endogenous chromophores. Polycylic aromatic hydrocarbons (PAHs) are a good example of exogenous chromophores. Light increases PAH impacts via photosensitization (e.g. generation of singlet oxygen) and by photooxidation of the chemicals to oxyPAHs. The modes of PAH action in solar radiation and the degree of involvement of oxyPAHs has been investigated extensively. OxyPAHs are toxic to plants, microbes and animals, and in many cases they are more toxic than the parent compounds. The site of cytotoxic action in plants is inhibition of photosynthetic electron transport. This has recently been shown to lead to synergistic toxicity between metals (e.g. Cu^{++}) and PAHs. The molecular sites of action are now being investigated by examining the proteins that are generated at very low PAH doses. Proteins act as endogenous UV chromophores in all organisms. A site of direct action of UV in plants is photosynthesis. We have been studying the effects of UV in natural sunlight on Lake Erie phytoplankton. Photosystem II (PSII) is being monitored by chlorophyll a fluorescence measured with a pulse-amplitude modulated (PAM) fluorometer. Sunlight exposures corresponding to surface irradiance under mid-day, clear sky conditions significantly impaired PSII activity in as little as 30 min. Both maximal PSII efficiency and steady state photosynthetic yield were strongly diminished, and the effects could be attributed to the UV in sunlight. Interestingly, the phytoplankton exposed to UV-B could not fully recover their ability to store energy after UV exposure.

215 Structural Photomodification Dramatically Changes the Environmental Fate and Toxicity of PAHs. B.M. Greenberg, D.G. Dixon, P. Mezey*, T.S. Babu, C.L. Duxbury, Y. El-Alawi, X.-D. Huang, A. Mallakin, C.A. Marwood, B.J. McConkey, and S. Tripuranthakan Dept. of Biol., Univ. of Waterloo, Waterloo, ON, Canada, *Dept. of Chem., Univ. of Saskatoon, Saskatchewan, SA, Canada. Modification of persistent contaminants (such as PAHs) is important for determining their fate and impacts. Photomodification (usually oxidation to oxyPAHs) changes the properties of PAHs, in many cases leading to more hazardous species. OxyPAHs have not been part of contaminant load assessments, even though photooxidation of PAHs is very rapid, with $t1/2$s in sunlight generally < 48 h. As well, several different quinones, phenols and carboxylic acids can be generated from each individual PAH. Thus, oxyPAHs are almost certainly contributors to total environmental PAH, and it is likely that real PAH impacts have been underestimated. We have determined the toxicity of several oxyPAHs, with many being more toxic than their parent compounds. This elevated toxicity of the photoproducts was observed with Lemna gibba, Daphnia magna, Photobacterium phosphoreum and fish, indicating a phenomenon that cuts across biological kingdoms. The mechanism of toxicity of oxyPAH photoproducts frequently involves inhibition of electron transport; respiration in animals and photosynthesis in plants. Next, we have computer modeled PAH phototoxicity using highly detailed electron-density shape-fragment maps of PAHs and oxyPAHs. With this information, a predictive model was trained based on the empirical toxicity data for several oxy- and intact-PAHs. The trained model should be able to predict the toxicity of a larger group of compounds. Finally, we have been assessing PAH contaminated aquatic samples from several sites to determine if oxyPAHs can be detected. The samples were analyzed by 2 dimensional HPLC. We have found modified PAHs in these samples. The implications of these results will be discussed.

216 Uv Photo-enhanced Toxicity of Carbaryl and Distilled Petroleum Product to Aquatic Organisms Little. E.E*., Hurtubise, R.D., Cleveland, L. USGS Environmental and Contaminants Research Center and A. Zaga, Wisconsin Department of Natural Resources. The potential interaction between ultraviolet radiation and environmental contaminants is seldom considered. Photolytic alteration of structure may increase or decrease the toxicity of the compound. Tissue damage may result from oxidation of tissue-bound contaminants by UV radiation. Studies were conducted to determine potential additive or synergistic effects of UV and potential environmental contaminants including a distilled petroleum product to a an estuarian fish, *Menidia beryllina;* and a pesticide, carbaryl, to tadpoles of the frogs, *Hyla versicolor* and *Xenopus laevis*. These substances are widely used and have a high probability of entering the environment. We conducted randomized tests using 4 to 6 toxicant concentrations and 2 to 3 UV irradiance intensities, alone and in combination. The 4 to 7 day static renewal exposures were conducted in 1 L beakers in a temperature controlled solar simulator that produced UVB, UVA, and visible light at environmentally relevant irradiance intensities. In the absence of UV, both substances were toxic only at the highest concentration (3.0 mg/L total petroleum hydrocarbon; 1.5 mg/L carbaryl). Exposures to UV alone were not harmful to the test organisms. However, the combined UV/ toxicant exposure resulted in significant mortality at all toxicant concentrations, even under limited UV exposures which amounted to less than 1% of the surface irradiance. The mode of photo-enhancement differed between the substances. The toxicity of petroleum was unaffected when the exposure solutions were irradiated prior to exposure, whereas UV exposure was lethal to fish that had been exposed to petroleum in darkness. This indicates that toxicity was induced by the oxidation of tissue-bound petroleum by UV radiation rather than by changes in the structure of the chemical. In contrast, fish previously exposed to carbaryl in the absence of UV were unaffected by subsequent UV exposure, whereas carbaryl solutions became significantly more toxic when they were irradiated prior to exposure. This indicates that UV-induced structural changes increased the toxicity of carbaryl. These data suggest that photoenhanced toxicity should be considered when evaluating the hazards of potential environmental contaminants, and also demonstrate that a range of aquatic habitats may be affected, given the low levels of irradiance needed to induce photoenhanced toxicity.

217 UV-B Exposure Increases Acute Toxicity of Pentachlorophenol and Mercury to the Rotifer *Brachionus calyciflorus*. Preston, B.L.*; Snell, T.W., School of Biology, Georgia Institute of Technology, Atlanta, Georgia; Kneisel, R., Dunwoody High School, Dunwoody, Georgia. Adverse biological effects of UV-B radiation have been well documented for phytoplankton and zooplankton in both marine and freshwater ecosystems. As UV-B is a ubiquitous environmental stressor, it has the potential to increase organisms' sensitivity to other stressors, thereby causing synergistic adverse effects. However, investigations of interactions between UV-B and anthropogenic toxicants have focused primarily on the chemical interactions between UV-B and the toxicant. Here we investigate the potential for UV-B to increase the sensitivity of the rotifer *Brachionus calyciflorus* to either acute pentachlorophenol (PCP) or mercury toxicity, independent of UV-B effects on these toxicants. *B. calyciflorus* neonates were exposed to sublethal UV-B of fixed irradiance for durations ranging from 10 to 60 minutes and then exposed to PCP or mercury in darkness for 24-hours in a standardized acute toxicity test. The effects of UV-B alone on *B. calyciflorus* reproduction and ingestion also were observed. UV-B exposures at the experimental doses had no effect on *B. calyciflorus* survival. However, UV-B increased the toxicity of PCP and mercury to *B. calyciflorus* as much five-fold, depending on duration of UV-B exposure and toxicant concentration. Reductions in the LC50 of up to 60% were also seen for both toxicants. UV-B alone effectively eliminated reproduction in *B. calyciflorus* and reduced ingestion by up to 90%. These results demonstrate the potential for UV-B to increase rotifer sensitivity to anthropogenic stressors independent of photochemical reactions with toxicants.

218 Influence of dissolved organic matter on phototoxicity of polycyclic aromatic hydrocarbons to grass shrimp and blue crab embryos. Lee,R.F.*, Kim,G.B., Maruya,K., Skidaway Institute of Oceanography, Savannah,GA; Steinert,S.A., Computer Sciences Corp., Marine Sciences Department, San Diego,CA. The toxicity of polycyclic aromatic hydrocarbons(PAHs) to aquatic biota increasures dramatically after exposure to sunlight. In our studies embryos from blue crabs,*Callinectes sapidus*, and grass shrimp, *Palaemonetes pugio*, were exposed for 24 hours in the dark to pyrene or anthracene at concentrations from 1 to 10μg/liter. After the dark exposure embryos were exposed to mid-day sunlight for periods of 1 to 4 hours. Embryos exposed to both sunlight (2 hours mid-day) and PAHs showed hatching effects with less than 20% of the embryos hatching at high PAH concentrations. Dark controls had 92 to 98% hatching success. A DNA strand damage (Comet) assay was used to determine the combined effects of sunlight and PAH exposure on embryo DNA. The comet assay measures the migration of fragmented DNA away from the nuclei of cells immobilized in agarose. The DNA was stained with a fluorescent stain and viewed using an epifluorescence microscrope. The distance and amount of DNA migrating from the individual nuclei was indicative of the number of strand breaks. Embryos exposed to PAHs and sunlight showed a high percentage of damaged DNA. There was some DNA damage after exposure to sunlight but significantly higher damage after exposure to both sunlight and PAHs. The influence of dissolved organic matter on PAH phototoxicity was assessed by adding aliquots of Ogeechee River water (dissolved organic carbon-50mg/liter) to seawater followed by addition of PAH and exposure to sunlight. High concentrations of dissolved organic carbon(10mg/liter) resulted in reduced PAH phototoxicity as determined by embryo hatching and DNA strand damage.

219 The Role of DOM in Phototoxicity to Aquatic Organisms. T. Mill, Chemistry Laboratory, Stanford Research Institute, Menlo Park, CA. Measurements confirm that short wavelength uv radiation is increasing owing to loss of stratospheric ozone, particularly in the Antarctic. Increased uv radiation increases both direct and indirect photoeffects on chemical species and organisms in the insolated water column, but the details and severity of phototoxic effects on organisms remain unclear. This paper explores the role of homogeneous and heterogeneous phototransient oxidants in oxidizing key biological components of aquatic microfauna. Transient oxidants formed by insolation of DOM include oxyradicals such as HO and RO_2, as well as singlet oxygen. Increased intensity of solar uv will increase rates of production of these photooxidants, their average concentrations (AC) and their rates of oxidation with reactive chemical bonds. HO has the greatest potential for biological effects because of its high reactivity toward almost any chemical bond; other oxyradicals and singlet oxygen are less reactive, however their AC are higher. There is good evidence that oxidant AC are controlled by dissolved organic components of the water column, not particulate or organism content. If photooxidation of a generic 2 mm diameter marine organism is modeled using a AC for HO of 1×10^{-17} M, and assuming a protein, polysaccharide or lipid wall, just 10% of the wall components will oxidize in about fifty days, much longer than the average organism's lifetime. However, if oxidant AC values are estimated from photoexcited DOM sorbed to cell surfaces, efficiencies of reaction are greater, even with the same AC, leading to higher rates of destruction of cell wall components. The extent of DOM absorption on aquatic cell surfaces is uncertain, but probably varies with the composition of the cell wall, pH and salt content of the aquatic system.

220 Effects of Ultraviolet Light And Polyaromatic Hydrocarbon Exposure on Sea Urchin Development And Bacterial Bioluminescence. Steevens, J.A.,* Slattery, M., Schlenk, D.K., Aryal, A., and Benson, W.H.,The University of Mississippi, University, MS. Polyaromatic hydrocarbons (PAHs) are relatively common contaminants of the Gulf of Mexico and may be activated to more toxic metabolites by ultraviolet light. Development of fertilized sea urchin (*Lytechinus variegatus*) embryos to the 4 and 32 cell stage was utilized as a measure of toxicity with co-exposures to Ultraviolet B (λ= 280-320 nm) light and the polyaromatic hydrocarbons, benzo[a]pyrene (BaP) and phenanthrene (Ph). Developing sea urchins demonstrated a significant ($p < 0.05$) dose dependent toxicological effect in response to exposure to BaP and Ph (1, 5, and 50 ppb), and UV-B light (5.8 uW/cm^2). Results of the sea urchin experiments were compared with a marine bacterial bioassay system which focuses on the reduction of luciferase-mediated bioluminescence in the microorganism *Vibrio fischeri*. Inhibition of bacterial bioluminescence was observed following exposure to naphthalene and phenanthrene (1, 5, and 10 ppm) for 5 and 15 minutes. UV-B significantly ($p < 0.05$) decreased bioluminescence in all treatments by 23.8 % as compared to control levels of light. No significant decrease in bioluminescence was observed with anthracene and BaP at concentrations up to 24 ppm and 1.6 ppm . Anthracene and BaP are relatively insoluble in water (less than 45 ppb), therefore, the bioavailability of the two PAHs may be a controlling factor of the exposure and effects to *V. Fischeri*. Interaction analysis demonstrates that concurrent exposure to UV-B and PAHs results in additive toxicological effects in both marine organisms tested. Results of the present study suggest that environmental conditions present in the Gulf of Mexico, including both UV light and PAH exposure, have the potential to cause adverse toxicological effects to aquatic marine organisms.

221 Dredged Material Evaluations - Risky Business? Moore, D.W.*, U.S. Army Corps of Engineers, Waterways Experiment Station, Vicksburg, MS; Cura J., Menzie-Cura & Associates, Inc., Chelmsford, MA. The current dredged material regulatory program is a tiered approach for determining the suitability of the material for disposal. It efficiently uses resources while ensuring that sufficient information is collected to make technically sound decisions. However, human health or ecological concerns sometimes arise that are not explicitly addressed within the existing tiered assessment framework. This presentation will: 1) review the concerns and conditions which the current framework does not sufficiently address; and 2) describe the Corps efforts to address these problems by integrating risk assessment approaches into the existing regulatory testing program. In each of the tiers data is collected to assess the potential for exposure and effects. Tiers I & II use existing information and simple screening tools while the later tiers use more sophisticated effects-based laboratory bioassays. In many cases, the interpretation of this information is obvious (e.g., toxicity in an acute sediment toxicity test renders the material unsuitable for aquatic disposal). However, in a smaller percentage of cases, interpreting the significance of the information is more difficult (e.g., slightly elevated tissue concentrations of a chlorinated organic observed during a bioaccumulation test). In addition, concerns sometimes arise that are not explicitly addressed within the existing tiered assessment framework (e.g., what is the risk to human health as a result of the potential trophic transfer of contaminants?). Questions such as these require a more thorough treatment of the information using approaches and methods common to environmental risk assessment. As a consequence the Corps is developing approaches for incorporating risk assessment into the existing regulatory testing program. This paper will describe a preliminary framework and discuss emerging tools to support risk-based assessments of dredged material.

222 Human Health and Ecological Risk Screen on the Proposed Open Water Disposal of Dioxin Contaminated Sediments for the Providence River Dredging Project. Tomey, D.A. US EPA Region I - New England, Boston MA. Risk screening for dioxin (sum of 2,3,7,8 TCDD toxic equivalents) was performed on the Providence River dredging project in support of EPA's approval of the open water disposal of this project's sediments. Bioaccumulation test results in clams, *Macoma nasuta*, after a 28 day laboratory exposure, were used to evaluate risk to: (1) human consumers of lobsters and finfish, and (2) developing finfish sac fry. The risk screen model assumed that consumed lobster and fish, and maternal fish were exposed to the clam after adjustment for steady state accumulation. Trophic transfer estimates (to lobster and fish tissues) employed octanol/water partitioning coefficients and known lipid values. For the human health screen, standard exposure and dose-response assumptions were utilized with modifications for recreational and subsistence fishers. We determined that the disposal of the tested sediments would not pose an unacceptable incremental risk for consumers of fish/shellfish taken from the disposal site area. To assess impacts to finfish, we estimated dioxin concentrations in fish eggs from maternal transfer. Predicted egg concentrations were below threshold values associated with adverse effects observed in developing lake trout sac fry. A review of the literature indicated that estuarine/marine fish were generally less sensitive than lake trout. We concluded that marine fish at the disposal site are not likely to be impacted by the levels of dioxin in the shellfish prey and, hence, the sediments.

223 Risk-Based Evaluation of Ocean Placement of Dredged Material Containing Dioxin. Peddicord, R. K., Dick Peddicord & Company, Inc, Parkton, MD. A predictive human health risk assessment was conducted to quantify the risks from dioxin associated with dredged material proposed for placement at the EPA-designated disposal site in the New York Bight apex. The food web model was tied directly to the steady-state dioxin bioaccumulation in polychaetes from the proposed dredged material, as calculated from the tests conducted as part of the regulatory evaluation of the dredging permit application. Dioxin was modeled in pelagic fish (bluefish), demersal fish (flounder) and shellfish (lobster). Human consumption was apportioned among these three types of fish, and between fish from the disposal site and fish not from the site. Consumption and risk were considered for recreational fishermen and for fish eaters obtaining their fish from the commercial catch (e.g., fish markets, restaurants). Exposures were calculated for a range of dioxin concentrations. For ease of use in the regulatory program, results were expressed in relation to polychaete dioxin uptake in the regulatory bioaccumulation test performed for the evaluation of dredging permit applications. Results demonstrated that under worst-case conditions, dredged material resulting in equilibrium polychaete body burdens of 140 pg/g dry weight or less (the equivalent of 10.85 pg/g wet weight as measured in the 28-day bioaccumulation regulatory test) could be placed at the EPA-designated dredged material disposal site off New York without exceeding 1 x 10 -6 risk. This is approximately 10 times the concentration considered acceptable under regulatory procedures in New York.

224 A Screening-level Environmental Risk Assessment for Comparison of Dredged Material Management Alternatives for NY/NJ Harbor. Kane Driscoll, S.B.,* Cura, J., Wickwire, W.T., Menzie-Cura & Associates, Inc. Chelmsford, MA; Moore, D.W., U.S. Army Corps of Engineers, Waterways Experiment Station, Vicksburg, MS. A screening-level ecological and human health risk assessment was conducted to compare relative potential risk among six dredged material management alternatives. Alternatives included disposal in subaqueous confined aquatic disposal pits (CADs), in island, near-shore or upland confined disposal facilities (CDF), in landfills, and the no-action alternative. A simple box model was used to predict exposure point concentrations in sediment and water surrounding the facilities. Results from actual bioaccumulation tests for highly contaminated, select NY/NJ sediments were used in a food chain model to predict daily doses and body burdens of contaminants in ecological receptors, such as invertebrates, fish and birds. Doses to human receptors, such as barge operators or fisherpersons, were calculated for uptake by ingestion, dermal contact, or inhalation. Seven criteria, based on the physical characteristics of each alternative, number of complete exposure pathways for human and ecological receptors, and the estimates of potential risk, were used to compare potential environmental impact among alternatives. The assessment identified complete exposure pathways that should be considered in future site-specific risk characterizations of these management alternatives. For example, accumulation of sediment-associated bioaccumulative compounds, food chain biomagnification, and trophic transfer to higher level consumers, such as piscivorous birds and fisherpersons is an important exposure pathway.

225 Surprising Dynamics During Ecological Recovery after Heavy Metal Contamination. Ferson, S.* Applied Biomathematics, Setauket, NY, Crutchfield, J., Carolina Power & Light, Raleigh, NC. The population of bluegill sunfish *Lepomis macrochirus* in part of a lake in North Carolina was decimated by toxicological and developmental effects of selenium leached from ash settling ponds. To forecast the potential recovery after cessation of heavy metal contamination, a demographic model was created for the bluegill population based on data collected from on-going biological monitoring at the lake. The model included density dependence which is known to be an important aspect of the life history of this species and used Monte Carlo methods to analyze the effect of natural environmental variability. The life history of the species revealed by analysis of the population model suggests that, if selenium poisoning were stopped, the population could recover to pre-impact abundances within two years. The increased abundance would be unevenly distributed among age groups, however. Following this increase in abundance, the biology predicts a population crash, especially among older year classes (which are prized by sportfishermen). This crash is due to the time-delayed effects of selenium on the population resulting from the strong non-linearity of density dependence in this species. The sharp increase in population size itself precipitates the crash. If this crash were not forecast in advance, its unanticipated occurrence could cause considerable consternation among managers, regulators and the interested public. This example shows that it can be important to predict ecological consequences to understand the nature and duration of biological recovery from toxicological insults. Without the understanding provided by the ecological analysis, the population decline would probably be completely misinterpreted as the failure of the mitigation program.

226 Using Sediment Quality Values in Ecological Risk Assessment of Dredging Operations. Chapman, P. M.*, EVS Consultants, N. Vancouver, B.C., Canada. Existing sediment quality values (SQVs) have various advantages but also a number of limitations. Their role in ecological risk assessment (ERA), considering both U.S. and non-U.S. ERA models, is reviewed, with particular emphasis on dredged material management. SQVs have three possible roles in ERAs: problem formulation, exposure assessment, risk characterization. Each of these possible roles is reviewed, and it is concluded that the only reasonable role for existing SQVs is in the problem formulation phase of an ERA. However, when and if research provides new SQVs which reliably provide cause-and-effect information, such SQVs may then be appropriate for more complex (and comprehensive) ERA phases. The reality that all SQVs are not the same is examined, together with the fact that there are no *best* SQVs, only *appropriate* SQVs, which can vary depending on site- and situation-specific factors. A simple model for using *appropriate* SQVs in a weight of evidence approach is proposed and then compared to guidance provided by the U.S. EPA and the ACOE for evaluating dredged materials, as embodied in the Green Book and Inland Testing Manuals. No change is required to use this model within the existing manuals.

227 Development of Sediment Quality Guidelines for the St. Louis River Area of Concern, Minnesota. Crane, J.L.*, Minnesota Pollution Control Agency, St. Paul, MN and MacDonald, D.D., MacDonald Environmental Sciences Ltd., Nanaimo, BC. The St. Louis River Area of Concern (AOC), in northeastern Minnesota, is one of 42 Great Lakes AOCs affected by contaminated sediments. Biologically-based sediment quality guidelines (SQGs) are being developed for this AOC in order to: 1) support the identification of contaminated sites, 2) prioritize contaminants of concern, 3) assist in the design, implementation, and evaluation of sediment quality monitoring programs, including trend analysis, 4) protect and/or restore healthy benthological habitats, 5) identify priority non-point source management actions, 6) design wetland restoration projects, 7) develop remediation clean-up goals, and 8) assist with the disposal and beneficial re-use of dredged material. A database of matching freshwater sediment chemistry and toxicity data for the St. Louis River AOC, Great Lakes region, and nationally (U.S./Canada) is being assembled. Site-specific SQGs will be developed for those chemicals having enough data to warrant their development. Otherwise, regional and national data will be used to augment the development of SQGs. An important objective of this project is to determine whether selected site-specific SQGs are statistically different from SQGs developed using regional and/or national data. An Advisory Committee is being used as a scientific sounding board for the development of the SQG approach. The following SQG approaches are being considered: logistic modeling, equilibrium partitioning, Apparent Effects Threshold (AET), Effects Range Low (ERL) and Median (ERM), and Threshold Effect Level (TEL) and Probable Effect Level (PEL). The SQGs derived for this project will be evaluated for assessments of comparability, reliability in the St. Louis River AOC, and predictability in other freshwater sediments. The utility of using SQGs in an ecosystem-based management approach will be discussed.

228 Towards More Ecologically Relevant Biological Testing in Contaminated Sediments: (I) Concordance of Benthic Bioassessment with Laboratory Chronic Bioassay Data under Washington's Sediment Management Standards. Chartrand, A.B., ENSR, Redmond, Washington. Biological testing and characterization of contaminated sediments is a major component of Washington's Sediment Management Standards (the Rule), but most frequently this testing is confined to standardized (acute and chronic) laboratory bioassay protocols. Benthic bioassessment, which relies on *in situ* measurements of infaunal communities to evaluate contaminated sediments, enjoys increasing acceptance and credibility in both the scientific and regulatory communities. In fact benthic bioassessment has been incorporated into the Rule as a component of biological characterization. In spite of the incorporation of both approaches into the Rule, little evaluation of concordance between these two very different approaches has been performed to date. This paper explores the respective benefits conferred by both approaches to the sediment characterization process, and the concordance between chronic laboratory bioassays and *in situ* benthic bioassessment, using accepted laboratory endpoints and benthic community metrics. Data were derived from specific case studies in which several types of synoptic data were collected (e.g. bulk sediment chemistry, pore water chemistry, bioaccumulation data, etc.). Such corroborating data were used to evaluate predictive capabilities of both kinds of biological data in detecting actual biological effects in contaminated sediments. Results of the comparison between the two approaches showed that chronic bioassays tended to be far more sensitive (conservative) than the native benthic communities, which are the organisms requiring protection. In addition, specific laboratory tests are generally more sensitive than others. For example, sediment larval tests tend to be more sensitive than other laboratory tests (e.g. chronic polychaete or amphipod somatic growth tests), which in turn are frequently more sensitive than benthic communities using commonly accepted metrics. This paper explores factors contributing to the general lack of concordance between these approaches.

229 OPPT Looks at the Contaminated Sediment Management Strategy: Why do we care? O. Meyn, J.Smrchek. United States Environmental Protection Agency. This presentation will discuss history and extent of sediment contamination from point and non-point sources in the United States and new approaches of management of these sediments. The document has been under development since 1992, is currently being revised based on public comments received in late 1994. The strategy introduces sediment quality criteria and provides a broad policy statement about use of criteria, commits EPA to use technically consistent sediment-assessment approaches, and provides a plan for reducing risks of ongoing and future contamination.

231 Quantification of Bioavailability of Heavy Metals for Plants and Oligochaete Worms. Baerselman, R., A.C. De Groot, D.T. Jager, W.J.G.M. Peijnenburg*,L. Posthuma, R.P.M. Van Veen, R.P.M., Laboratory for Ecotoxicology, National Institute of Public Health and the Environment, Bilthoven, The Netherlands. Soil particulate phases frequently have high concentrations of metals relative to the concentrations of metals in the dissolved phase. Total concentrations of contaminants, however, are not always directly related to effects since exposure of biota may be limited by processes that render the metals unavailable for uptake. The aspect of differences in availability among different soil substrates has only to a limited extent been included in the present approaches for deriving ecotoxicity-based soil quality criteria. In this contribution, results are presented of a study aimed at providing a scientific basis for inclusion of availability in assessing ecological risks of polluted sites. It is assumed that each biotic species can be considered as one of the soil phases next to the particulate phase and the liquid phase. Dynamic equilibrationprocesses are assumed to take place between all phases present. The *biotic phase* consists of a variety of species, each with a characteristic set of exposure routes. The equilibrium partitioning concept was used in our study as the basis for describing the physico-chemical equilibria outside the organisms. Partition coefficients of metals differ strongly among 20 moderately metal-contaminated field soils as a function of soil characteristics. Thereupon, time-dependent uptake of metals by three plant species and by two oligochaete worms was studied in the 20 soils. Uptake and elimination characteristics and bioconcentration factors for these species are shown to be related to the same soil characteristics that determine soil-water partitioning. This indicates that uptake of metals by the organisms studied is either directly from the pore water or from both the pore water and an uptake route that is related to the pore water.

232 Simultaneous Measurement of Uptake and Depuration of Trace Metals Using Stable Isotope Tracers. Evans, R.D.*, Balch, G., and Welbourn, P.W., Trent University, Peterborough, ON, Canada. The accumulation of contaminants in organisms is the net result of the opposing processes of uptake and depuration. To properly understand or model the kinetics of trace metal accumulation both rate constants must be measured. Traditionally each parameter is measured separately, often on different organisms. Of particular concern is the fact that frequently the depuration is measured at near-zero contaminant concentrations; a situation which is not reflective of natural conditions. We have developed a method which allows the simultaneous determination of both uptake and elimination constants using stable isotopic tracers. Organisms are exposed to one isotope for a period of time (typically 5 days) following which the first isotope is removed and they then are exposed to a second isotope. During this second phase it is possible to determine both uptake and loss on the same individuals simultaneously. Moreover, the elimination during the second phase is determined with contaminant concentrations at steady-state. We have used this method for determining uptake and loss constants for Cd, Cu, Ni, Pb and Zn in *Hydropsychidae* (caddisfly larvae) held in flow-through containers. Experiments were conducted using sub-part per billion concentrations that reflect typical non-contaminated systems. Using this system uptake and elimination are measured easily, even at exposure concentrations as low as 100 parts per trillion. The results indicate that the kinetics of the various metals are quite different. Cadmium and zinc behave similarly with rapid uptake and depuration, while lead is released much more slowly. Results using our approach will be compared to those from companion depuration experiments conducted in water containing no contaminant.

233 Modeling Uptake of Ag, Cd, and Zn by the Estuarine Amphipod *Leptocheirus plumulosus* from Multiple Uptake Routes. Schlekat, C.E.*, Chandler, G.T., Decho, A.W., University of South Carolina, Columbia, SC. Because benthic invertebrates directly consume metal-contaminated surficial sediments, these organisms represent the limiting factor that controls the flux of the vast repository of metals from sediments to upper level wildlife. To further understand processes controlling metal transfer from sediments to benthic invertebrates, we used a toxico-kinetic approach to model the uptake of Ag, Cd, and Zn by the estuarine amphipod *Leptocheirus plumulosus*. *L. plumulosus* lives infaunally and feeds facultatively on suspended material and surficial sediments. The model incorporated these components: 1) metal-particulate partitioning, 2) metal uptake from dissolved and dietary routes, 3) feeding rates, and 4) metal depuration from both uptake routes. Metal-particulate partitioning, which dictates the proportion of metal available through each uptake route, was determined across a broad salinity range (2.5-25‰) between each metal and a variety of environmental particles, including two types of organic carbon sediment coatings, a mineralogical sediment feature, and two species of phytoplankton. Dissolved uptake was measured directly, whereas the efficiency with which *L. plumulosus* assimilated metals from each particle type was used, in conjunction with feeding rates, to estimate dietary uptake. Results show that *L. plumulosus* should accumulate a substantial proportion of total Cd and Zn through dietary ingestion routes because these metals partition strongly to particles and because they are assimilated by amphipods with relatively high efficiency (approximately 20%). Ag, which does not strongly partition to particulate matter, should be rapidly accumulated via dissolved uptake routes. Finally, uptake of both Ag and Cd is influenced by salinity, which affects both partitioning and dissolved uptake. In summary, this model shows that *L. plumulosus* should accumulate metals at a higher rate under oligohaline estuarine conditions.

234 The Bioavailability of Mercury to the Estuarine Amphipod *Leptocheirus plumulosus*. Lawrence, A. L.* and Mason, R. P. Chesapeake Biological Laboratory, University of Maryland, Solomons, Maryland. The bioaccumulation of inorganic mercury (Hg) and monomethylmercury (MMHg) by benthic organisms directly influences the amount of metal transferred up the estuarine food chain and is difficult to predict. Three routes of exposure must be examined when assessing the potential exposure of amphipods to mercury: exposure from water or porewater, sediment, and food. Microcosm laboratory experiments were performed to determine the major route of mercury uptake. Hg and MMHg were analyzed using standard analytical techniques for low-level measurements and quantified by atomic fluorescence. The bioavailability of sediment bound Hg and MMHg was evaluated with regard to acid volatile sulfur and sediment organic matter content, which varies significantly among coastal sediments. Hg and MMHg are particle reactive and have a strong affinity for organic matter, a potential food source for amphipods. Amphipods living in organic rich sediment spiked with MMHg and Hg accumulated less mercury than those living in sediments with a lower organic matter content, but the same metal concentration. Experiments designed to investigate the bioavailability of MMHg and Hg from ambient seawater with varying amounts of organic matter showed that in the presence of equal metal concentrations, amphipods living in water with little or no organic matter accumulated more mercury than those living in water with a greater percentage organic matter. MMHg was more readily available for uptake than Hg in all exposures. Bioconcentration factors (BCF) were used to assess the relative importance of the uptake routes; sediment and food were more significant than overlying water and porewater. BCFs from these experiments are in agreement with field data from the Chesapeake Bay and Lavaca Bay, TX.

235 Modeling and Measuring Metal Accumulation in Marine Copepods. Fisher, N.S.*, W.-X. Wang, I. Stupakoff, S. Sanudo-Wilhelmy, J.-L. Teyssié, S.W. Fowler, and J. Crusius. Marine Sciences Research Center, SUNY Stony Brook, Stony Brook, NY. To field-test predictions of a kinetic model regarding metal concentrations in marine zooplankton, we analyzed Cd, Zn, Ag, Co, and Se in surface water, phytoplankton, and zooplankton samples collected off the coast of Monaco. Mean dissolved metal concentrations in surface waters from 10 stations were 10.5 + 2.4 pM for Ag, 72.3 + 16.2 pM for Cd, 260 + 43 pM for Co, and 4.3 + 1.6 nM for Zn. Kd values for suspended particles were not affected appreciably by particle load and were on the order of 1 e5 for Co, 3 e4 for Ag, 1 e4 for Zn, and 8 e3 for Cd. Mean metal concentrations in copepods (nmol/g dry wt) were 1.3 + 0.4 for Ag, 1.3 + 0.8 for Cd, 9.5 + 2.9 for Co, 16.0 + 3.8 for Se, and 2570 + 830 for Zn. These metal concentrations were close to concentrations predicted by a bioenergetic-based kinetic model which used experimentally determined assimilation efficiencies, influx rates from food and water, and efflux rates. This is the first field verification of a kinetic model of contaminant bioaccumulation in marine plankton. The close fit of predicted and measured values suggests that the model takes into account all the major factors which influence metal concentrations in these animals. Further, the model predicts that Se and Zn are obtained primarily from food and Ag, Cd, and Co are obtained about equally from food and water.

236 Bioavailability of Metals from Sediments to a Deposit-feeding Polychaete. Wang, W-X.*, Department of Biology, HKUST, Hong Kong, and Fisher, N.S. Marine Sciences Research Center, SUNY, Stony Brook, NY. Assimilation efficiencies (AEs) of trace elements (Ag, Cd, Co, Se, Hg, methylmercury, and Zn) in the marine deposit-feeding polychaete *Nereis succinea* from ingested sediments were measured using radiolabeled encapsulated sediments. The ranges of AEs were 12-36% for Ag, 5-44% for Cd, 35-96% for Co, 7-30% for Hg, 29-60% for Se, and 21-59% for Zn. AEs of $CH_3Hg(II)$ ranged between 43 and 83% and were strongly affected by sediment composition. AEs of other metals were little affected by sediment organic carbon content and grain size. Metals (but not methylmercury) in anoxic sediments were assimilated with a lower efficiency than metals from oxic sediments. The AE of Cd decreased, and the AE of Co increased, with the duration of sediment radiolabeling; AEs of Ag, Se and Zn were weakly affected by sediment aging. Metal uptake in worms from the dissolved phase was generally proportional to concentration in the dissolved phase. Uptake rate constants were highest for $CH_3Hg(II)$, followed by Ag > Hg > Zn > Cd > Co > Se. $CH_3Hg(II)$ accumulation contributes about 5-17% of total Hg accumulation in polychaetes. A bioenergetic-based kinetic model, which incorporates metal influx from both the dissolved and particulate (sediment) phases, indicates that most (>98%) of the Cd, Co, Se and Zn in polychaetes is due to ingestion of sediment because of the high ingestion rates of these animals and the low uptake rate of metals from the dissolved phase (porewater or overlying water). For Ag and Hg, an estimated 5-35% is due to uptake from the dissolved phase. Our study suggests that sediment quality criteria must consider sediment as a potentially important source for metal uptake in benthic invertebrates.

237 Effects of AVS (Acid Volatile Sulfide) on the Bioaccumulation of Cd, Ni, and Zn in Bivalves and Polychaetes. Lee, B.-G.*, Jeon, H.-S. Luoma, S.N., USGS, Menlo Park, CA; Yi, J.-S., Koh, C.-H., Seoul Natl Univ. S. Korea. Laboratory microcosm experiments were conducted to investigate the influence of acid volatile sulfide (AVS) on the bioaccumulation of Cd, Ni, and Zn in two estuarine clams, *Potamocorbula amurensis* and *Macoma balthica*, and two ploychaetes, *Heteromastus filiformis* and *Neanthes arenaceodentata*. In eight treatments, animals were exposed to four levels of Cd (0.02 ~ 2 μmole/g), Ni (0.6 ~ 6 μmole/g) and Zn (2 ~ 15 μmole/g) in sediments with a constant AVS (6 μmole/g), and four AVS levels (0.3 ~ 30 μmole/g) in sediments with a constant metal concentration for 18 d. The influence of AVS on metal bioaccumulation was determined by biological attributes of the organisms such as feeding strategy and burrowing habits, and was compounded by the geochemical factors such as oxidation of surface sediments and co-variance with SEM. Following exposure, tissue Cd, Ni and Zn in both clams increased linearly with SEM, except for Ni in *P. amurensis*. Bioaccumulation by the clams did not follow either SEM/AVS ratio or porewater metals. The clams appeared to accumulate metals from ingested oxidized surface sediments. Accumulation of Cd in the surface burrowing worm *Neanthes arenaceodentata* was strongly related to both the SEM/AVS ratio and porewater Cd; but bioaccumulation of Ni and Zn was related only to SEM. Accumulation of Ni in the headdown deposit feeder *Heteromastus filiformis* was not influenced by AVS but by SEM. *Heteromastus filiformis* did not accumulate significant amounts of Cd and Zn.

238 Dietary Availability of Metals Incorporated into Aquatic Invertebrates: Are Species and History of Metals Exposure Important? Suedkamp, M.J., University of Wyoming, Laramie, WY; Farag, A.M., U. S. Geological Survey, Jackson, WY; Meyer, J.S., University of Wyoming, Laramie, WY. We rinsed six invertebrate diets at pH 2 and 7, and analyzed concentrations of four metals (Cd, Cu, Pb and Zn) and total protein in the rinsates. Four diets were prepared from macroinvertebrates collected from metals-contaminated rivers; the other two diets (one of which was exposed to metals in the laboratory) were prepared from laboratory-reared *Artemia*. To further investigate associations between metals and proteins, the pH 2 rinsates were fractionated using size-exclusion chromatography (SEC). Metals and protein leachability differed considerably among several of the diets. In pH 2 rinsates, approximately 4X more protein was leached from the metals-free *Artemia* diet than was leached from all other diets. A significantly larger percentage of the total Cu in both *Artemia* diets was leached at pH 7 than in two of the macroinvertebrate diets, whereas similar percentages of total Cu were leached from the *Artemia* diets and the same macroinvertebrate diets at pH 2. In SEC fractions from both *Artemia* diets, but not from any macroinvertebrate diets, Cu and protein co-eluted. None of the other three metals co-eluted with protein in any of the diets. These results suggest fundamental differences in metal binding properties and protein leachability between invertebrate diets. Thus, different invertebrates and different histories of metals exposure may lead to different metals and protein availability to predators.

239 a Cross-phyletic Comparison of Contaminant Bioavailability. Weston, D.P.*, University of California, Berkeley, CA; Mayer, L.M., University of Maine, Walpole, ME; Gravitz, L., University of California, Berkeley, CA; Bock, M., University of Maine, Walpole, ME. *in vitro* digestive fluid extraction is useful to quantify the bioavailability of sediment-bound contaminants. The sediment of concern is incubated *in vitro* in digestive fluid of a deposit-feeding invertebrate, and the proportion of contaminant solubilized is considered as the maximum amount that would be bioavailable for absorption during gut passage. Most past work has been with arenicolid polychaetes, but in this study we extend the approach to 19 species of invertebrates representing 7 phyla. Digestive bioavailability of benzo[a]pyrene (BaP) and zinc from sediment varied by about 10-fold depending on the species from which the digestive fluid was obtained. The potential for contaminant solubilization by all species was far below that extracted by organic solvents or acid as might be used in a chemical analysis, indicating that standard extraction techniques do not reflect bioavailability. The potential for contaminant solubilization was related to phylogenetic similarity. Digestive fluid from four species of arenicolid polychaetes extracted roughly similar amounts of BaP (15-25%) and Zn (10-15%). Echinoderms, both holothuroids and echinoids, tended to have digestive fluids that were very poor at solubilizing hydrophobic toxicants or zinc and, in general, were comparable in extraction efficiency to seawater. Fluids from echiurans and priapulids tended to be among the most effective extractants (25-40% BaP, 15% Zn). These relationships are interpretable based on fluid characteristics include enzyme activity, organic carbon and lipid content, and surfactant properties. These data clearly indicate that the concept of "the bioavailable contaminant fraction" is highly species-specific, and dependent upon phylogenetically-based differences in gut fluid characteristics.

240 The Role of Dissolved Metal Binding Ligands on Metal Speciation in Marine Sediments. J. Shine. Harvard School of Public Health, Boston, MA. Research has shown that the activity of contaminants in porewater may be related to their bioavailability to benthic organisms. For metals, the free ion concentration in porewaters may be an appropriate species predicting biological availability. Research has been appropriately directed at examining the role of sulfides (acid volatile sulfides) and particulate organic carbon as binding phases reducing the free ion concentration of metals in the porewater. Less work has been done examining the extent that dissolved organic ligands in porewater may contribute to reduction of free metal ion concentrations. As part of a larger study to refine our knowledge of metal speciation and bioavailability in marine sediments, the role of dissolved organic ligands on free copper ion concentrations in pore water was examined in contaminated sediments from New Bedford Harbor, USA, and 'clean' control sediments from Buzzards Bay, USA The free metal ion content of the pore water was examined using an acetylacetonate competitive ligand technique to determine the concentration and conditional thermodynamic stability constants of the metal binding ligands. The results indicate two classes of dissolved ligands: a strong ligand with a log conditional stability constant of approximately 12, and a weaker ligand with a log conditional stability constant of approximately 10. In general, the binding capacity of the dissolved ligands is on the order of milli-equivalents/liter. This is the same order of magnitude as the concentration of acid volatile sulfides. Speciation model results indicate that the dissolved copper binding ligands can make a significant contribution to reduction of copper activity in sediments, and thus may contribute to reduction of bioavailability.

241 Test Methods to Determine Environmentally Acceptable Endpoints of Chemicals in Soil Based on Availability to Groundwater and Human and Ecological Receptors. Linz, D.G.*, LinzTech, Inc., Des Plaines, IL; Nakles, D.V., Remediation Technologies, Inc., Monroeville, PA; Harju, J., Gas Research Institute, Chicago, IL. The Gas Research Institute is conducting an extensive research initiative to develop a technical protocol for determining environmentally acceptable endpoints, or EAEs, for chemicals in soil, based on their availability to groundwater and to ecological and human receptors. This research initiative includes both laboratory and field investigations to identify the extent and mechanisms of sequestration of chemicals in soil, the rate and extent of chemical release to groundwater, the rate and extent of uptake by ecological and human receptors, and the fate of chemicals in the subsurface environment under natural environmental conditions. The specific objective addressed in this paper is the development and evaluation of test methods for field soils that can be used to define risk-based concentration limits for a chemical based on its availability to the relevant pathways (groundwater, ecological or human receptor). The results have shown that chemicals in field soils, especially chemicals present in mixtures in soils that have been aged and weathered in the environment, are not completely and immediately released to the environment or receptors, and therefore total concentration measurements will over-predict the risk associated with these soils. This behavior in aged and weathered soils is evidenced by: 1) reduced leaching by water and other solvents, 2) reduced mineralization by microbes, 3) reduced toxicity to plants, earthworms, and insects, 4) reduced uptake in mammals and reduced dermal absorption. Factors that have been identified include the soil texture and surface area, soil organic matter content and quality, mineral matter nanoporosity, environmental conditions such as aging time and wet/dry cycles, and the presence of a residually trapped nonaqueous phase liquid (NAPL). This work has defined a set of measurements (bioassays, chemical assays, leaching tests, membrane/sorbent tests, *in vitro* dermal absorption tests) that can be used to determine the availability of chemicals from soil, and to utilize this availability data to define acceptable soil concentrations based on environmental risk. The results show that bioassays and chemical assays can be correlated, and that surrogate measurements could potentially be used for soil regulatory determinations.

242 Determining Environmentally Acceptable Endpoints for PAH-Contaminated Soils. Harkey, G.A.*, Cepuritis, A.M., Srivastava, V.J., Institute of Gas Technology, Des Plaines, IL. Bioavailability of PAHs was examined in archived soils from a number of Manufactured Gas Plant (MGP) sites. Data on physical and chemical properties of these soils gathered during previous treatability studies was collected from various reports and databases. The soils were further characterized for contaminant bioavailability via extraction studies with mild solvents and toxicity tests using earthworms (*Eisenia foetida*) and bacteria (Microtox® tests). The four main objectives of this ongoing project were to 1) Establish a database containing treatability information on selected projects completed or in progress for determination of Environmentally Acceptable Endpoints (EAEs) for contaminated soils; 2) Add toxicity data to such a database for use in tracking toxicity with treatment over time; 3) Correlate toxicity with the amount of contaminants extracted using mild organic solvents; and 4) Verify the accuracy of the Microtox® Solid-Phase test for its use in predicting soil toxicity. Toxicity data obtained from untreated MGP-site soils using earthworm and Microtox tests showed significant differences in bioavailability of PAHs, depending on the soils examined. These results showed no correlation between the total PAH concentration in soils as obtained from traditional chemical/physical extraction methods and toxicity. For example, one soil with a high concentration of total PAHs (2694 ppm) showed 100% earthworm survival after 14 days and a Microtox EC50 of greater than 1% dry weight of soil, a value considered to be relatively non-toxic. Another soil with a concentration of 660 ppm total PAHs yielded 0% earthworm survival after 7 days and a Microtox EC50 of 0.04%.

243 Effects of Soil Organic Matter Geochemistry on the Desorption and Bioavailability of Soil- and Sediment-Associated Phenanthrene. Walter J. Weber, Jr., Weilin Huang, Zhi Dang, Angela Lueking, Subhasis Ghoshal Department of Civil and Environmental Engineering, The University of Michigan, Ann Arbor, MI 48019-2125. Rates of desorption and mineralization of phenanthrene presorbed on four different geomaterials were measured independently. Tenax polymer beads were used to enhance the desorption process and a Pseudomonas cepacia CRE7 strain was used in the mineralization studies. Both rates and extent of phenanthrene desorption and mineralization were found to be functions of the geochemistry of the organic matter associated with the geomaterials. Lachine shale, a geologically old and diagenetically altered material containing highly reduced and condensed organic matter, was found to exhibit slow desorption and mineralization rates. More than 50% of phenanthrene associated with this sample was neither readily desorbed nor available for microbial mineralization within an incubation time of 30 days. Conversely, Chelsea soil, a geologically young and moderately humified material containing both condensed and amorphous organic matter, exhibited a biphasic desorption process; a fast desorption phase followed by a slow but persistent release. The majority of the phenanthrene sorbed by this material was readily available for mineralization within the 30 day incubation period. It appears that both desorption and bioavailability are constrained by slow diffusion of solute within condensed soil organic matter matrices. The study suggests that a valid scientific rationale exists for establishing alternative endpoints for soil/sediment remediation.

244 Testing an Earthworm Test, for Assessing the Biological Quality of Soils: What Works, and Why? Stewart*, A. J., Wicker, L. F., Nazerias, M.S., Merchant, S. D. and Short, D. K. Oak Ridge National Laboratory, PO Box 2008, Oak Ridge, TN. We studied effects of soil quality and test conditions on growth and reproduction of the earthworm, *Eisenia foetida*, using a 21-d test method described previously. We assessed 19 soil samples from various petroleum-industry sites; 10 of the soils contained total petroleum hydrocarbon (TPH) residuals, and 9 (reference soils) were nearly TPH-free. Soils with higher levels of TPH were tested using dilutions, made with a contaminated site's corresponding reference soil. Earthworm growth and reproduction data generally did not relate well to soil TPH level, but we found reasonably good concentration - response relationships for growth in three TPH-contaminated soils. We used experiments to explore effects of testing conditions on *E. foetida* growth and reproduction, in two reference soils (dark vs constant light, 1 g vs 5 g of food per replicate, and fresh vs aged food), and we conducted two experiments to see if earthworm water-content was a significant source of error in estimates of earthworm growth. Overall, we found that soil type and test conditions can strongly influence *Eisenia* growth and reproduction, even in reference soils. But when test factors are accounted for properly, earthworm growth (and perhaps reproduction) can yield evidence for sublethal effects of pollutants.

245 Effect of Extraction Method in Determining Toxicity in PAH-Contaminated Soils. Harkey, G.A.*, Cepuritis, A.M., Institute of Gas Technology, Des Plaines, IL; Young, T.M., University of California, Davis, CA. Extractability of PAH-contaminated soils as a function of extraction method was studied, using Microtox® tests. Soils from five former Manufactured Gas Plant sites were extracted via Soxhlet, supercritical carbon dioxide (SC CO_2), and saline methods, using standard procedures (e.g., ASTM, USEPA). Microtox Solid Phase tests were conducted on soils prior to extraction and after SC CO_2 extraction, while Microtox Acute tests were conducted on samples from SC CO_2 and saline extractions. No correlation between total PAH concentrations via Soxhlet or SC CO_2 extractions and EC50s was seen, although toxicity was lower in soils after extraction, except for one case where no change in toxicity was apparent. Toxicity of saline-extracted samples was often consistent with negative controls and was not considered toxic for any of the soils examined. Microtox tests performed on samples from SC CO_2 - extracted soils showed significantly greater toxicity in fractions that contained less volatile PAHs, compared to more highly volatile PAH fractions. The enhanced toxicity seen with samples extracted by vigorous chemical/physical means provides evidence that unnatural extraction procedures can strip sequestered contaminants from soil particles that would normally be unavailable for uptake to biological receptors.

246 Use of Ion Exchange Membrane Probes to Assess Potential Ion Supply Rates in Soil Environments. Schoenau, J.J., University of Saskatchewan, Saskatoon, SK, Canada. Ion exchange resins in membrane form can be used to assess supply rates of inorganic and organic ions in samples of soil and directly in the field. The approach we developed involves use of cation and anion exchange resin membranes encapsulated in plastic probes. The probe is inserted directly into the soil where it is allowed to remain for a period of up to two weeks. During this time, elements that are released into available ionic forms exchange with counter ions on the resin membrane surface and become sorbed. Measurement of sorbed ions on the resin membrane surface provides a indication of potential ion supply rate under relatively undisturbed soil conditions. This technique has been used to study bioavailability of nitrate, phosphate, metals, organic acids and herbicides in soils and sediments. These studies included soils receiving different rates of fertilizer, manure, and herbicide applications as well as soils contaminated by heavy metals. We have found supply rates of inorganic and organic ions measured in this matter to be as well and often better correlated with plant uptake and response than concentrations determined using chemical extracting solutions.

247 Modeling of PCB Biodegradation: Evaporation, Distribution, and Mapping of the First Oxygenase Binding Site. Balaz, S.*, North Dakota State University, Fargo, ND; Vrana, B., Hornak, V., Dercova, K., Slovak Technical University, Bratislava, Slovakia. The kinetics of distribution of PCB and other hydrophobic chemicals in soil and aqueous media is governed by evaporation, adsorption to solid particles and biomass, and by microbial degradation. Many published degradation rate constants are imprecise because all the processes were not considered in their determination. The published relationships between structure and degradation rate constants of individual PCB congeners are of surprisingly low quality taking into account limited conformational flexibility, inertness and inability of the compounds to ionize. One of possible causes is that the symmetrical biphenyl skeleton facilitates multiple orientations (binding modes) of PCB molecules in the binding site. A complete kinetic description of the fate of individual PCB congeners in the suspension of *Pseudomonas stutzeri* as determined by structure and physicochemical properties of the congeners and of the biomass has been developed. Monitoring of evaporation and distribution allowed for determination of the degradation rate parameters. The parameters were correlated with the structure and properties of the congeners by the receptor mapping procedure considering multiple binding modes. Conformational energy and conformational entropy of the congeners were included in the expression of the free energy of binding to the first oxygenase. The approach provided a reliable structure-degradation relationship.

248 Canadian Soil Quality Guidelines for Petroleum Hydrocarbons. Gaudet, C.*, Ouellet, S., Gagnon, C., Environment Canada, Ottawa, Ontario; Nason, G.E., Alberta Environment, Lethbridge, Alberta; Gagnon, C., Schneider,T., Environmental Consultant, Ottawa, Ontario. In 1997, The Canadian Council of Ministers of the Environment (CCME) requested development of soil quality guidelines for petroleum hydrocarbons to lend national consistency to the assessment and remediation of petroleum contaminated soils in Canada. Based on the results of a multi-stakeholder workshop held in Canada, the CCME recommended a tiered approach to the development of soil quality guidelines for protection of the environment and human health, consistent with the Canadian Framework for Assessment and Remediation of Contaminated Sites: i.e. Tier 1 generic guidelines, Tier 2 site-specific objectives, and Tier 3 risk assessment. Options for Tier 1 guidelines are based on elements of the existing British Columbia Environment (BCE) and Total Petroleum Hydrocarbon Criteria Working Group (TPHCWG) approaches, and define petroleum hydrocarbons in terms of specified carbon fractions. Based on an initial evaluation, the CCME proposes that Tier 1 be based on 3 to 4 windows or carbon ranges representing various molecular weights (volatile, light extractable, heavy extractable, and possibly higher MW in the $>C_{32}$ to C_{50} range). At the Tier 1 level, it is also recommended that 3 (or 4) guidelines be derived distinct carbon boiling point ranges with no separation of the alkanes/cycloalkanes and aromatics/alkenes. Currently, three multi-stakeholder Technical Advisory Groups (TAGs) have been set up to advise the CCME on precise delimiters for the recommended carbon ranges, the need for a fourth heavier fraction ($>C_{32}$ up to C_{50}), surrogates for each boiling point range fraction; and plausible Tier 2 and 3 approaches.

249 Ecotoxicologically Based Soil Quality Criteria - Protection of the Terrestrial Communities Against Hazardous Substances. Throl, C., Federal Environmental Agency, Berlin, Germany. Until now there are no soil quality criteria for the protection of the terrestrial communities in Germany. In contrary there is a concept for the protection of surface inland waters, which encloses the protection of the aquatic communities. In analogy a concept for the derivation of soil quality criteria is developed, which allows a systematic and transparent derivation of reference concentrations for hazardous substances in soils. Bellow these reference concentrations in soils no adverse effects on terrestrial communities are expected. The concept take into account effects of chemicals through the effect routes soil → microflora, soil → soil organisms, and soil → birds/mammals feeding on soil invertebrates. For selected metals and organic substances soil quality criteria are deduced. Applying the equilibrium partitioning approach, the effect data are a) normalised for a standard soil (OECD soil) and b) extrapolated for the three main kinds of soils clay, loam/silt, and sand. For each effect route is deduced a criteria on the basis of the lowest effect date. The lowest of the route specific criteria forms the soil quality criteria for a selected substance. The results are compared with the precautionary values of the German soil protection act and soil quality criteria of other countries.

250 Metal Standards for Fertilizer and Supplement Products in Canada: Derivation of the Standards, Background Information and Calculations. Webster, L.G.*; Blair, D.H., Canadian Food Inspection Agency, Government of Canada, Nepean, On, Canada. In 1980, the Fertilizer Section established federal standards for acceptable levels of metals in sewage sludge. These were derived from standards for maximum cumulative metal additions to soils. The standards for maximum additions were based on the existing Ontario provincial guideline for sewage sludge and information regarding the background levels of metals in soils across Canada. The following points were considered during the development of the guidelines and standards: it was recognized that safe metal concentrations for soils had not been established; average metal concentrations in soils had been determined via research investigations; a number of factors, in addition to the metal content of the soil, affect the uptake of metals by plants; it appeared that the concentration of metal in the plough layer was the most appropriate standard for the limits on metal addition; and the maxima for recommended metal content of soils and metal addition to soils were based on the best judgement of a number of people familiar with the literature on metals. The Ontario and federal standards compare well, with the federal ones being slightly higher for certain metals. It is important to understand that the standards are predicated on the maximum, acceptable, cumulative metal additions to soils, and not a maximum acceptable metal concentration in the product per se. The application rate of the product, and hence the accompanying metals, is a crucial aspect of the use of the standards for any given product. Two examples will be used to demonstrate the application of these standards.

251 Phytochelatins as Biomarkers for Heavy Metal Toxicity in Plants from Natural Vegetations. Sneller, F.E.C.*, Van Heerwaarden, L.M., Schat, H., Verkleij, J.A.C. and Ernst, W.H.O. Vrije Universiteit Amsterdam, The Netherlands. Phytochelatins (PCs) are heavy metal-binding peptides, specifically produced in plants upon exposure to heavy metals. They have the general structure (γ-glu-cys)$_n$-gly, where n=1-11. Recent recearch in our laboratory showed a strong correlation between short term toxicity of Cu and Cd and PC-production, independent of metal-tolerance (Schat and Kalff, '92, De Knecht *et al.*, '94). Laboratory and field experiments were carried out to test the possibility to use PCs as biomarkers for heavy metal toxicity. Under laboratory conditions, PC concentrations were strongly correlated with short term As-toxicity and long term Cd-toxicity. It was shown that in field situations, PCs could be detected in various plant species from sites contaminated with As and Cd. A bioassay with the several test species, varying in metal sensitivity, is currently developed. We conclude that in the case of exposure to strong inductors of PC production, such as Cd and As, PCs can give be used as specific biomarkers of metal toxicity. In the case of Zn, which is a very poor inductor, PCs do not reflect toxicity. Another biomarker will have to be developed to assess the toxicity of this element. Cited literature: Schat, H. and Kalff, M.M.A. (1992) Are phytochelatins involved in differential metal tolerance or do they merely reflect metal-imposed strain? *Plant Physiology* 99, 1475 - 1480. De Knecht, J.A., Van Dillen, M., Koevoets, P.L.M., Schat, H., Verkleij, J.A.C. and Ernst, W.H.O. (1994) Phytochelatins in cadmium-sensitive and cadmium-tolerant *Silene vulgaris*. Chain length distribution and sulfide incorporation. *Plant Physiol.* 104, 255 - 261.

252 Metal Body Burden in Sentinel Organisms as a Quantitative Biomarker of Ecological Impact. Birge, W.J.*, Hogstrand, C., Shaw, J.R., and Price, D.J., University of Kentucky, Lexington, KY. Metal body burden (*e.g.* Ag, Cd, Cu, Ni, Pb), particularly when used in conjunction with metallothionein (MT) and MT-mRNA, may provide an integrated real-time quantitative biomarker system for predicting ecological effects of metals. In bioassessment studies of a 2nd to 3rd order stream affected by low concentrations of several metals, several invertebrate and piscine species were evaluated as sentinels of metal exposure. Metal body burden was greatest in metal-sensitive mayflies; intermediate in stoneroller minnows (SM); and least in metal-tolerant caddisflies. Indicator species have proven useful in evaluating effects of pollution and this investigation indicates that metal body burden is a useful parameter in identifying metal-sensitive to metal-tolerant taxa. SM, a benthic herbivore, was selected as the principal sentinel monitor for this study. This species is moderately tolerant to metals and, in this study, regulated whole body burden proportional to exposure. Metal body burden (*e.g.* Ag, Cd, Cu) varied among SM from eight different monitoring stations, giving close inverse correlation ($r \geq -0.95$) with ecological conditions, including species richness, abundance and Protocol III scores for macroinvertebrates. Correlation coefficients (r) for metal body burden *vs* the EPT index ranged from -0.82 for Ag to -0.94 for Cd. SM grazes on periphyton and integrates exposure over time, spatial distribution, and from all avenues of exposure. It is useful in characterizing sources of bioavailable metal and body burden values may be used to calculate metal multipliers for application to total recoverable metal concentrations. Strong inverse correlations (r) were obtained for calculated bioavailable silver and copper concentrations *vs* number of macroinvertebrate taxa (-0.95 Ag, Cu), bioassesssment scores (-0.97 Ag; -0.99 Cu), and the EPT index (-0.82 Ag; -0.87 Cu).

253 Unique protein signatures in *Mytilus edulis* exposed to copper, PCBs and decreased salinity. Shepard, J.L.*, UMBC, Baltimore, MD; Olsson, B.; Tedengren, M. University of Stockholm, Stockholm, Sweden; Bradley, B.P., UMBC, Baltimore, MD. The entire complement of proteins produced by an organism, the proteome, is now being studied extensively in relation to human health and functional genomics. In environmental toxicology these well developed techniques from proteome analysis may be used to identify chemical-specific protein signatures of exposure in tissues from any organism. It has already been well documented that protein profiles differ among stressors to which the organisms or cells are exposed. The aim of this project was to verify that specific protein signatures could be identified in mussels exposed to both anthropogenic and natural stress and that these signals could also be identified in mussels exposed to more than one stressor. *Mytilus edulis* from the Baltic Sea were brought into the laboratory at the University of Stockholm Asko marine laboratory and subjected to treatment with copper (70ppb), Arochlor 1248 (1ppb), salinity lowered from 6ppt to 3ppt and to lowered salinity plus copper exposure. Eight mussels per treatment group were acclimated in the laboratory for 24 hours before beginning the seven day exposure. At the end of the exposure period mussels were removed from the tanks, snap frozen and freeze dried for shipment to the University of Maryland (BC). Whole body tissue was then homogenized and seperated using two-dimensional gel electrophoresis. Protein gels were then scanned to Tiff files and analyzed using MELANIE II 2D gel analysis software (BioRad[a]). Protein signatures including proteins both induced and suppressed by exposure were identified for each treatment group and compared. Proteins induced and suppressed by all treatments were identified as well as proteins induced and suppressed by each treatment. Unique portions of both the copper and salinity signatures could be identified in the salinity plus copper treatment group. Protein signature analysis using the techniques developed for proteome analysis can successfully be used to identify exposure of aquatic organisms to specific stressors and simple mixtures of toxicants.

254 The Effects of *in vitro* Paraoxon Exposure on Brain and Muscle Acetylcholinesterase Activity in Two Estuarine Fish Species. Fulton, M., Key, P., NOS, Charleston, SC; Andrews, M., Princeton University, Princeton, NJ. Organophosphorus insecticides (OPs) are among the most widely used pesticides worldwide. These compounds produce toxicity by inhibiting the enzyme, acetylcholinesterase (AChE), which leads to an accumulation of the neurotransmitter, acetylcholine, that ultimately causes a blockade of nervous impulses. Previous work in our laboratory has shown that the relationship between OP-induced AChE inhibition (in brain and muscle tissue) and mortality is quite different for two estuarine fish species, the red drum *(Sciaenops ocellatus)* and the mummichog *(Fundulus heteroclitus)*. In the red drum the 24h EC_{50}s for brain and muscle inhibition were 5.2 μg/L and 6.8 μg/L, respectively, and were quite similar to the 96h LC_{50} of 6.2 μg/L. In contrast, the 24h EC_{50} for brain AChE in the mummichog was 1.0 μg/L while the 96h LC_{50} was 64.5 μg/L. No effect on muscle AChE was observed in the mummichog at 5.0 μg/L. The goal of this study was to evaluate the *in vitro* sensitivity of brain and muscle tissue from each of these species to paraoxon (a direct AChE inhibitor) to determine if differences in target site sensitivity might explain the results observed *in vivo.* Tissue samples were incubated with a range of paraoxon concentrations and I_{50} values calculated. Muscle AChE activity was more sensitive to paraoxon-induced AChE inhibition than brain tissue in each species. Mummichog brain tissue was more sensitive than brain tissue from the red drum. There was no species-related difference in muscle tissue sensitivity. These results suggest that some, but not all of the differences observed in the *in vivo* studies may be related to target site sensitivity.

255 Comparison of innate immune parameters from *Oncorhynchus mykiss* caged at various sites in the Hamilton Harbour. Karrow, N.A[1].*, Bennie[2], Boermans[3], H. J., Bols, N.C.[1], Brown, S.[2], Dixon, D.G.[1], Gamble, A.[2], Ganasson, R.[1], Parrott, J.[2], Solomon, K.R.[3], & Sherry, J.[2]. [1]University of Waterloo, Waterloo, ON, [2] Environment Canada, National Water Research Institute, Burlington, ON, [3] University of Guelph, Guelph, ON. A preliminary field study was conducted in the Hamilton Harbour to determine whether or not exposure, at sites known to be contaminated with high levels of polycyclic aromatic hydrocarbons (PAHs) and heavy metals, was sufficient to alter fish immune function. Caged fish (*Oncorhynchus mykiss*) were sampled after 7, 14, and 21 days of exposure from six sites designated by their proximity to highly contaminated sediments and various industrial and municipal discharges. Pronephros leukocytes were evaluated for phagocytic activity, oxidative burst, and surface immunoglobulin-positive B cell counts. Serum was collected to monitor lysozyme levels. All immune parameters were compared to fish sampled from a Lake Ontario control site. Both the phagocytic index and percent of phagocytizing leukocytes were reduced across time in fish sampled from the harbour sites compared to the Lake Ontario control fish; oxidative burst was also reduced at two of these sites. B cell counts were not significantly affected across time at any of the sites, however, overall fish B cell numbers at three of the harbour sites appeared to be lower than the Lake Ontario controls. Serum lysozyme levels were also not affected across time, but were significantly reduced at one of the harbour sites. These preliminary results indicate that innate immune parameters are altered in fish caged at various sites within the harbour. Some of those alterations also appear to be time dependent.

256 Biochemical Markers of Exposure to Crude Oil and Their Relationships to Tainting in Fish. Gagnon, M.M., RMIT, Melbourne, Australia; Holdway D.A., RMIT, Melbourne, Australia. Australian crude oils are classified as light crude oils with a large portion being water soluble. Therefore, toxicity of these crude oils may be greater than observed for other heavier oils. To assess toxicological effects of an Australian crude oil, selected biochemical markers of exposure were validated using Atlantic salmon *(Salmo salar)* exposed to Bass Strait crude oil. Variations in biomarkers were followed during a 6-day exposure to the water accommodated fraction of crude oil or to chemically dispersed crude oil, and during a 31-day depuration period. Induction of ethoxyresorufin-O-deethylase (EROD) measured in the liver demonstrated rapid uptake of soluble compounds, while fluorescent metabolites present in the bile confirmed metabolism and excretion of petroleum compounds. Disturbances in the carbohydrate metabolism was evidenced by altered levels of the aerobic enzymes citrate synthase and cytochrome C oxydase, and of the anaerobic enzyme lactate dehydrogenase. All biomarkers measured can potentially be used to monitor exposure and depuration of salmon stocks to petroleum compounds, while only the most sensitive biomarkers can be used as commercial tools to ascertain absence of petroleum compounds in the flesh of pre-exposed fish.

257 Cytochrome P450 Monooxygenase Activities as a Biomarker for Pcb Exposure and Effect in Field Collected and Manually Dosed Tree Swallow (*Tachycineta bicolor*) Nestlings. Yorks, A.L.*, Melancon, M.J., and Hulse, C.S., USGS Patuxent Wildlife Research Center, Laurel, MD; Stegeman, J.J., Woodin, B.R., Woods Hole Oceanographic Institute, Woods Hole, MA. The utility of measuring protein and catalytic function of cytochrome P450s of the CYP1A and CYP2B subfamilies as biomarkers for environmental polychlorinated biphenyl (PCB) exposure in upland birds was examined using Tree swallows (*Tachycineta bicolor*). From 1995 to 1997, nests and nestlings were monitored at eight sites in Maryland, Pennsylvania, and New York. Additionally, eggs and nestlings from one site were dosed with the model cytochrome P450 inducers, β-naphthoflavone, 3-methycholanthrene, PCB 126, and various Aroclors. Hepatic microsomes were prepared from twelve-day old nestlings and analyzed for EROD (CYP1A) and BROD (CYP2B) activities. A subset of microsomal samples were also examined by protein immunoblotting using polyclonal antibodies specific for the CYP1A and CYP2B subfamilies. Heart and skin samples from the nestlings were examined immunohistochemically to determine relative amounts and cellular location of CYP1A. Although BNF induced cytochrome P450s as assessed by enzyme activity and protein immunoblotting, PCBs were ineffective. A range of PCB contamination was seen in composite sediment (0.02-16ppm) and egg (0.70-9.7ppm) samples from the field sites. BROD and EROD activities correlated significantly with each other ($r>0.8$) and with PCB concentrations. Contaminant levels and CYP1A and CYP2B antibody responses of the nestlings also were correlated. Differences between sites and treatments show that there is a cytochrome P450 biomarker response in nestling Tree swallows which is indicative of contaminant exposure and effects. However, lack of enzyme activity after dosing with PCBs indicates that induction at field sites may be due to other contaminants.

258 Chorioallantoic Membrane Use as a Non-lethal Indicator of Avian Exposure to Organochlorines, a Laboratory Validation Study with White Leghorn Chickens (*Gallus domesticus*). [1]Bargar, T.A., [1]Taylor, M.D., [2]Cobb, G.P., [1]The Department of Environmental Toxicology, Clemson University, Pendleton, SC, 29670. [2]The Institute of Human and Environmental Health, Department of Biological Sciences, Texas Tech University, Lubbock, TX, 79409. A study was performed to investigate the relationship of PCB and endosulfan levels in the chorioallantoic membrane (CAM) with toxicological endpoints in White Leghorn (*Gallus domesticusl*) hens and chicks. Experiment one investigated the influence of lipophilicity and chemical interaction upon chemical excretion into the egg. Adult hens were injected with congeners 105, 156, 189 and endosulfan mixed isomers. Yolk and albumin from each egg were separately extracted and analyzed. Lipophilicity influenced chemical excretion into the yolk. Significantly greater amounts of congener 105 (3943 ng) were excreted into the yolk relative to congeners 156 (3422 ng, $p=0.077$) and 189 (2367 ng, $p=0.0001$). Experiment two investigated the relationship of increased dose to hens with chemical levels in the CAM and toxicological endpoints (P450 activity and steroid hormone levels). Adult hens were subcutaneously dosed with corn oil, 0.65 $\mu g/\mu l$, 1.34 $\mu g/\mu l$ and 2.0 $\mu g/\mu l$ corn oil. Dosing compounds were the same as in experiment one. Hens were artificially inseminated following one week of dosing. Ten eggs were collected from each hen following insemination initiation. Eggs 1, 3, 5, 7, and 9 were incubated until hatch while eggs 2, 4, 6, 8, and 10 were saved for extraction and analysis. Preliminary results indicate that chemical levels in the CAM are correlated with dose to the adult hen and toxicological endpoints (P450 activity) in both chicks and hens. This study shows that the CAM can serve as a non-lethal indicator of avian exposure to organochlorines.

259 Effects of Tnt, Rdx and Hmx on Growth, Reproduction, Immune Function, Glutathione Status and Various Enzymatic Biomarkers in *Eisenia foetida*. Inouye, L.S.*, ASCI Corporation, Vicksburg, MS; McFarland, V.A., Waterways Experiment Station, Vicksburg, MS; McCant, D.M., ASCI Corporation, Vicksburg, MS. The earthworm, *Eisenia foetida*, was utilized to determine potential biological effects of three explosives (2,4,6-trinitrotoluene (TNT), octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX), and hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX)). Earthworms were exposed to artificial soil spiked with TNT (~200 ppm), RDX (~400 ppm), or HMX (~400 ppm) for a 21 day period. Both RDX and HMX reduced growth (weight gain) and reproductive output (cocoon production). All three compounds impacted immune function by either decreasing the number of coelomic leukocytes (coelomocytes) or nitroblue tetrazolium dye reduction by coelomocytes (an assay for nonspecific immune function). Total glutathione concentrations decreased upon exposure to TNT, while exposure to HMX resulted in an increase in total glutathione concentrations and exposure to RDX caused no significant alterations on this parameter. Activities of glutathione peroxidase (selenium dependent and selenium independent), glutathione reductase, superoxide dismutase, catalase, and glutathione S-transferase were also measured. Exposure to TNT caused an increase in catalase activity and a decrease in selenium independent glutathione peroxidase activity, while both RDX and HMX caused a decrease in glutathione reductase activity. Overall, growth, reproduction and immune function give the most consistent results and appear to be the most sensitive assays.

260 Testing for a Bottleneck Event in Central Stoneroller (*Campostoma Anomalum*) Fish Populations Inhabiting the Tributaries of the Great Miami River Basin P. Dimsoski, Oak Ridge Institute of Science and Education, G.P. Toth, S.A. Christ, R.N. Silbiger, and M.K. Smith, NERL, United States Environmental Protection Agency, Cincinnati, OH. A Monte Carlo based sign test was conducted to test for the reduction of the effective population size (i.e. a bottleneck event) in the recent history of six populations of the Central Stonerollers inhabiting the tributaries to the Great Miami River. Six sampling sites with ten fish per site were randomly selected from twenty sites in that basin being studied as part of a U.S. EPA Regional Environmental Monitoring and Assessment Program (REMAP) effort. The DNA extraction from the tissue of fish was followed by PCR amplification in which six pairs of microsatellite primers were used. The amplified products were separated by capillary electrophoresis. Allele frequencies were used to calculate the expected heterozygosity, as well as the heterozygosity observed under mutation-drift equilibrium for both infinite allele and stepwise mutation models. The results of the test, using both models, showed that in four of the six populations, two had heterozygosity excess for five loci and two for six loci which supports the scenario of a recent reduction of population size. The data support the hypothesis that the present genetic variation observed for the four populations of Central Stoneroller in the Great Miami River basin is primarily the result of a recent bottleneck which reduced the population size, rather than due to natural selection acting over a longer period of time. Though the power of this test can be increased by adding more individuals and loci, these findings should be taken into consideration when genetic variation is interpreted in the context of vulnerability studies for the ecoregion.

261 Organotin Compounds in Biota in the Waters Between Sweden and Denmark. Jacobsen, J.A.*, Ministry of Environment and Energy, National Environmental Research Institute, Department of Marine Ecology and Microbiology, Denmark; Strand, J., Institute of Biology and Chemistry, Roskilde University, Denmark. Tributyltin (TBT) used as toxic ingredient in antifouling paints on ships is expected to be widely distributed in the marine environment. Biota was collected from 15 stations in the narrow and busy shipping straits between Sweden and Denmark. Organotin species, originating primarily from antifouling paints on ships, were extracted by in-situ derivatisation followed by CGC-DC-PFPD determination. Samples obtained were Buccinum undatum, Neptunea antiqua, Arctica islandica, Musculus niger, and Neptunea antiqua. Butyltin concentrations in body tissue from deposit feeders were higher than in suspension feeders. TBT wet weight concentrations were between 1 and 82 ng /g ww (as Sn) n= 35 samples whereas none was below limit of detection of around 0.5 ng/g ww (as Sn). The concentration levels and the ratio between TBT and degradation products was highly species dependent. Gastropods tend to transform TBT to DBT better than mussels. The distribution of butyltin compounds in the Sound region shows trends towards the major shipping ports($p<0.01$) This study indicates a marginal significant trend across shipping lanes($p<0.06$). The deposit feeding mussel Nuculana pernula is an ideal monitoring organism for horsiontal distribution of butyltin compounds in these waters.

262 Trends in Environmental Concentrations of Tributyltin Compounds in Coastal Waters off Europe and Japan. Läenge, R., Schering AG, Berlin, Germany. The use of tributyltin (TBT) compounds in antifouling paints has been limited to commercial shipping in the previous years due to legislative regulations. In monitoring programs the concentrations of TBT were studied in order to investigate the influence of commercial shipping on TBT levels as well as the effect of the regulations on the reduction of TBT in coastal areas. In European monitoring programs performed in the early 90s, the trends show a very significant decrease of TBT in mooring and marina areas that have been contaminated by small boats. Related to commercial shipping, harbors with docking and shipyard activities were predominant sources for TBT contamination. There was no indication that shipping lanes in coastal areas were a main source for water column contamination by TBT. In Japan monitoring of water column, sediments, and biological tissue since the late 80s indicated a rapidly decreasing trend in all areas monitored, and most samples show now TBT concentrations at or below the detected limit (3 ng/l) in the water column.

263 Contamination of Organochlorine and Butyltin Compounds in Deep-sea Organisms Collected from Japanese Coastal Waters and the Western North Pacific. Takahashi S.*; Lee, J.S.; Tanabe, S., Ehime University, Matsuyama, Japan; Kubodera, K., National Science Museum, Tokyo, Japan; Kawaguchi, K., Ocean Research Institute, The University of Tokyo, Tokyo, Japan. The role of deep-sea bed has been considered as a final reservoir of persistent contaminants. Despite this, very few studies have examined contaminant levels and their behavior in deep-sea ecosystem. In the present study, organochlorines, such as PCBs, DDTs, CHLs, HCHs and HCB, and butyltin residues including TBT and its breakdown products were determined in deep-sea organisms collected from Suruga Bay, a deep coastal water body in Japan, and the western North Pacific. Concentrations of organochlorines such as PCBs and DDTs in deep-sea organisms from Suruga Bay appeared to be lower than those in deep-sea organisms reported in 1970~80s. Whereas these organisms accumulated significant concentrations of butyltins at levels comparable to those reported in fish from Tokyo Bay and other coastal waters in developed countries, suggesting the expansion of butyltin pollution in deep-sea ecosystems around Japan. Among butyltin derivatives, TBT was the predominant compound in most deep-sea organisms, suggesting a continuous input of TBT into the deep-sea environment and lower metabolic capacity to degrade TBT by these organisms. With regard to mesopelagic myctophids from the western North Pacific, relatively high concentrations of PCBs, DDTs and CHLs were observed in species living deep waters, whereas HCHs, HCB and butyltins were accumulated at higher concentrations in species migrating shallower water. These patterns found in deep-sea organisms might have been influenced by the horizontal and vertical distributions of organochlorines and butyltins in the western North Pacific waters. To our knowledge, this is a first report on the detection of organotin residues in deep-sea organisms.

264 Assessment of Chronic Risks to Aquatic Organisms Posed by Tributyltin. Brancato, M.S.*, Cardwell, R.D., Tear, L., McKay, S., Parametrix, Inc., Kirkland, Washington. Chronic risks posed by tributyltin (TBT) to generic aquatic communities and sensitive marine species were evaluated using probabilistic techniques. Risks to aquatic communities from TBT were assessed in different matrices. The risk assessments were consistent with the U.S. EPA's risk assessment framework and with a method developed by the Water Environment Research Foundation (WERF). The WERF method fits Probability Density Distributions (PDFs) to TBT concentration and effects data for multiple species. The exposure and effects PDFs are then integrated to estimate the percent aquatic species affected. The results suggested that the most sensitive species to TBT may be at risk. Thus, effects to these sensitive species (e.g., Mercenaria mercenaria, Ostrea edulis, Crassostrea gigas, Acartia tonsa, and Nucella lapillus) were evaluated further. Generalized Linear Models (GLIMs) were fitted to literature data to estimate dose-response relationships for each of these species. Model coefficients were used to calculate percent reductions in survival or growth, relative to controls, expected over a range of TBT concentrations. Using probabilistic risk assessment techniques and information about historical and current TBT concentrations at different sites, we estimated past and current risks to individual organisms. Extrapolation from individual to population-level risks requires age-specific survivorship and reproduction rates for individual populations. Because survival, growth, and reproductive rates can vary greatly depending on environmental conditions, we constructed several different generic Leslie matrices and varied these life history rates to evaluate potential effects on population growth rates.

265 Use of the Imposex and Intersex Indicators of Tributyltin (TBT) Contamination in Eastern Canada. Prouse, N.J.*, Bedford Institute of Oceanography, Dartmouth, N.S., Canada; Covert, T.J., Halifax Fisheries Research Laboratory, Halifax, N.S., Canada. Tributyltin (TBT) biocides used in marine antifouling paints cause masculinization of female marine gastropods (snails). The resulting abnormalities, termed *imposex* (development of penis and vas deferens in female neogastropods) and *intersex* (modification of female genital tract towards a male system in littorinids) are used as sensitive indicators and biological monitors of TBT contamination. A baseline survey of eastern Canada in 1995 found intertidal dogwhelks (*Nucella lapillus*) affected by imposex at 13/34 sites examined. Since 1995, new sites have been examined and other indicator species included. Periwinkles (*Littorina littorea*) inhabiting rocky intertidal shore were severely affected by intersex near areas of high boating activity, e.g. sites in Halifax Harbour. Imposex was observed in edible whelks (*Buccinum undatum*), a subtidal neogastropod of potential commercial importance, sampled from Halifax Harbour and approaches. Mud snails (*Nassarius obsoletus*) found on mudflats were imposexed in several harbours. Concentrations of TBT and other organotins in tissues and sediment were measured and related to biological effects. Sites are being resurveyed and the intensity of imposex or intersex measured to monitor any changes in TBT-induced effects. Results suggest TBT is a widespread problem particularly in large vessel harbours.

266 Determination of Realistic Leach Rates for Environmental Risk Assessment. Arias, S., Elbro, H.S., and Kjaer, E.B., Hempel Marine Paints A/S,Lyngby, Denmark; Graziano, L.C., Jacobson, A., McGuckin, T.A., and Willingham G.L., Rohm and Haas Company; Spring House, PA; Nichols, C, Rohm and Haas Company, Croydon, UK. In order to determine the environmental safety of new biocidal antifouling coatings it is necessary to develop models which will predict the environmental loading incurred by their use. Risk assessments generally follow the risk paradigm, *Risk is a function of hazard and exposure*. The hazard function is obtained from toxicity testing data. The exposure function is composed of degradation rates, environmental partitioning, and inputs to the environment. Degradation rates and environmental partitioning under a range of conditions can be accurately determined experimentally. Inputs to the environment, however, are more difficult to estimate in the laboratory with the same degree of accuracy. Since the environmental input is the most sensitive factor in the calculation of PEC/PNEC (Predicted Environmental Concentration / Predicted No Effect Concentration), it is essential that an accurate value is employed. In this presentation, we shall compare various approaches to estimating environmental input (laboratory and field studies and calculation methods), discuss their relative validity, and their use in environmental risk assessment.

267 Comparisons of Risks to Aquatic Life from Using Tin-free Biocides Versus Tributyltin in Antifouling Paints. Brancato, M.S., Toll, J.*, Keithly, J., Cothern, K., Kluck, M.J., Henderson, D. and DeForest, D., Parametrix, Inc., Kirkland, WA. Tin-free biocidal additives are currently being promoted as alternatives to tributyltin (TBT) for use in antifouling paints. As part of a cost-benefit analysis on antifouling paint additives, we found a significant cost penalty to the shipping industry if currently available tin-free biocides were used and TBT paints were banned. Given this, it is important to compare the environmental risks of tin-free biocides and TBT self-polishing copolymer paints. TBT risks to aquatic life have dropped as water concentrations have declined in all regions of the United States since OAPCA. Unfortunately, the toxicity data for alternative biocides are for acute exposure durations, and therefore unsuitable for assessing antifouling paint risks (because antifouling paints are continuously leached from hulls). Despite the data shortcomings, we have been able to draw some conclusions about the potential environmental risks associated with tin-free alternative antifouling paints containing Sea-Nine 211 or Irgarol 1051. Using modeled environmental concentrations, we derived a lower 95 percent confidence limit on the fifth percentile acute toxicity threshold of 0.026 μg/L. Using this threshold, we found acute toxicity from Sea-Nine 211 to aquatic life at five of six modeled sites (Antibes, Golfe Juan, Norfolk Harbor, San Diego Bay, and a hypothetical Dutch harbor). These results compare unfavorably to TBT, which has not exceeded acute toxicity thresholds in U.S. waters since 1992. The data for Sea-Nine 211 and Irgarol 1051 illustrate that one cannot assume tin-free antifouling paints are as safe as or safer for the environment than TBT SPC paints.

268 Ecological Risk Assessment for Irgarol 1051: an Algaecide for Antifoulant Paints. Hall, L. W. Jr*., University of Maryland, Queenstown, MD; Giddings, J. M., Springborn Laboratories, Wareham, MA; Solomon, K. R., University of Guelph, Guelph, Canada; Balcomb, R., Ciba Specialty Chemicals Corporation, Tarrytown, NY. Irgarol 1051 is an algaecide used in copper-based antifoulant paints for controlling fouling organisms on the hulls of recreational and commercial watercraft. Paints containing this algaecide have been used in Europe since the mid-I 1980's; however, recently these paints have been registered for use in the United States. To examine the risk that Irgarol may pose to aquatic ecosystems, a probabilistic ecological risk assessment was conducted using a distribution of exposure and toxicity data. Exposure data for this assessment were derived from 11 monitoring studies (146 stations) conducted in marinas, ports, estuaries and coastal waters from 1992-1997 in six European countries. A comparison of 90th percentile values pooled by station types across all regions showed that values in marinas and small ports (317 and 447 ng/L) were higher than in estuaries and coastal waters (41 and 19 ng/L). A 90th percentile of 133 ng/L was reported for all pooled stations. Temporal trends showed that Irgarol concentrations typically peaked in early summer after launching of small boats with much lower values occurring during the spring, fall and winter. Toxicity data used for this risk assessment were derived primarily from unpublished studies submitted to regulatory agencies. Since Irgarol is a herbicide, it is much more toxic to plants than animals. Therefore, a conservative approach using a distribution of plant toxicity data was used to derive a 10th percentile of 135 ng/L based on LC/EC5O data. Results from probabilistic analysis showed that ecological risk from Irgarol exposure was generally judged to be low at most locations.

269 Global Environmental Risk Assessment of Sea-nine™ 211 Biocide, a New Active Ingredient in Marine Antifouling Coatings. Kramer, V.J.* and Jacobson, A., Rohm and Haas Company, Spring House, PA. Novel marine antifouling agents will be required in the event that current proposals to eliminate tin-based antifoulant paints are enacted. Sea-Nine™ 211 Biocide is a new active ingredient for marine antifouling coatings that has been proven to be effective in limiting the growth of biofouling organisms on marine vessels. An environmental risk assessment of the use of Sea-Nine™ 211 Biocide as the active ingredient in a marine antifouling coating was conducted. Harbors in the United States, Europe and the Asian Pacific region were modeled using a variety of scenarios and modeling tools that were considered appropriate for each region. An extensive freshwater and saltwater toxicity database on Sea-Nine™ 211 Biocide was used to estimate conservatively the concentration in the marine environment expected to cause no adverse impact on aquatic organisms. Species sensitivity distribution analysis as well as assessment factor approaches were used to calculate no-effect concentrations. Hazard quotients were calculated for each harbor scenario. The rapid biotic and abiotic degradation of Sea-Nine™ 211 Biocide in the marine environment is predicted to result in concentrations well below toxicity thresholds. Therefore, when used in accordance with manufacturer's recommendations, Sea-Nine™ 211 Biocide in marine antifouling coatings is expected to cause no adverse effects to non-target marine organisms. Sea-Nine™ 211 Biocide can be considered a safe alternative to tin-based antifouling agents.

270 From Anti-dandruff to Antifoulant: Zinc Pyrithione, a Non-persistent Alternative to Tbt. Turley, P. A.*, Audette, D.E., Fenn, R.J., Ritter, J.C. Olin Biocides Technology, 350 Knotter Drive, Cheshire, CT 06410 Zinc pyrithione (ZPT) is an effective bactericide, fungicide and algaecide with a long history of safe usage. It was introduced over 30 years ago under the trademark Zinc OMADINE®* as an antidandruff agent and has become the leading antidandruff additive in shampoos. ZPT has broad spectrum activity and when added to marine paint, is found to be a highly effective antifoulant in combination with cuprous or zinc oxide. The negative impact of tributyltin (TBT) on some aquatic and marine organisms has highlighted the importance of assessing environmental fate and persistence of antifoulants. We have carried out several studies of ZPT in fresh and salt water systems to determine degradation rates and the pattern of formation and decline of degradates. These studies were designed to meet the requirements of the United States Environmental Protection Agency for registering antifoulants and include leach rates, abiotic hydrolysis and photolysis, aerobic and anaerobic aquatic metabolism, adsorption/desorption and die-away studies. The findings are consistent with ZPT's history of usage and the absence of ecological effects. ZPT degrades rapidly in the water column by both abiotic and biotic pathways to products that are orders of magnitude less toxic than ZPT. Although pyrithione adsorbs to sediment, facile cleavage of a critical functional group precludes accumulation in anaerobic sediment. ZPT environmental fate data and physical property data were used to calculate predicted environmental concentration (PEC), persistence and distribution. TBT data, obtained from the literature, were similarly treated. The results of exposure modeling strongly suggest that ZPT is far less persistent, particulary in sediment, and is environmentally more favorable than TBT.

271 Detection of Estrogenic Chemicals Using Vitellogenin-specific Rt-pcr. Selcer, K.W.*, Duquesne University, Pittsburgh, PA; Williams, L.K., Duquesne University, Pittsburgh, PA; Palmer, B.D., University of Kentucky, Lexington, KY. A number of chemicals released into the environment mimic the action of the steroid hormone estrogen. These environmental estrogens pose potential health risks to humans and wildlife by disrupting physiological and developmental processes. Our laboratories are designing *in vivo* bioassays for environmental estrogens based on induction of vitellogenin in the African clawed frog, *Xenopus laevis*. We have previously assessed estrogenicity using immunological assays for serum vitellogenin. In the present study, we have developed reverse transcriptase polymerase chain reaction (RT-PCR) assays for hepatic vitellogenin mRNA induction as a measure of estrogenicity. Using *Xenopus laevis* vitellogenin-specific primers, the RT-PCR procedure generated the expected size cDNA fragment (761 base pairs) with hepatic RNA from frogs immersed in the estrogenic agent diethylstilbestrol (DES, 1 ppm, 11 days), but generated no product with RNA from control frogs. The cDNA fragment was cloned into a plasmid, sequenced, and found to have 100% identity with the *Xenopus laevis* vitellogenin cDNA sequence. The RT-PCR method was dose-responsive with regard to total hepatic RNA added, from 0.25 to 2.0 μg DES-treated frog RNA . Vitellogenin mRNA was detected within 4 h after immersion in DES and reached maximal levels at 24 h. DES-induced vitellogenin mRNA induction was dose-responsive from 1 ppm to 0.01 ppb. Vitellogenin mRNA was detected at low levels in all frogs immersed in the weakly estrogenic compound nonylphenol, and in half of frogs immersed in p-octylphenol. The present research demonstrates the utility of RT-PCR for detection of *Xenopus laevis* hepatic vitellogenin mRNA. The vitellogenin mRNA induction assays developed could be used to screen chemicals for estrogenic properties or to test waters for the presence of estrogenic agents. Supported by NIH ES07621 and USDA 9504243.

272 Validation of a Short-term, Whole Embryo Assay to Evaluate Adverse Effects on Amphibian Metamorphosis and Thyroid Function Using *Xenopus Laevis*. Fort, D.J., Propst,T.L., Schetter, T., and Stover, E.L., The Stover Group, Stillwater OK. Short-term static-renewal studies were performed on *Xenopus* embryos with pentachlorophenol, nonylphenol, methopreneand methimazole (anti-triiodothyronine [T_3]), from day 50 (Stage 60) to day 64 (Stage 66) [14-d test] to evaluate effects on tail resorption and thyroid function. Pentachlorophenol and methimazole decreased the rate of tail resorption significantly in metamorphs, yielding no adverse effect level NOAEL) and lowest adverse effects level (LOAEL) values of 5.0 and 10.0 μg/L and 10.0 and 25.0 μg/L, respectively. Methoprene completely arrested metamophosis when embryos were exposed to a concentration of 5 μg/L during stages 8-46 (day 0-4). No effects were detected, however when methoprene was administered from stages 60-66 (day 50-64). Nonylphenol increased the rate of tail resorption, yielding NOAEC and LOAEL values of 25.0 and 50.0 μg/L, respectively. Because combined exposure to thyroxin and pentachlorophenol did not appreciably increase the rate of tail resorption, pentachlorophenol appeared not to be acting by thyroid disruption, but by some other mechanism, possibly by compromising bioenergetics. Addition of thyroxin to 5 μg/L methoprene treated embryos from day 0-4 did not alter the rate of tail resorption indicating as with pentachlorophenol that some other mechanism was being affected. The stimulatory effects observed with nonylphenol appeared to be associated with thyroid activity since the rate of tail resorption was decreased to a rate similar to the control values when nonylphenol was tested in combination with methimazole. The *Xenopus* model appeared to be a suitable system for evaluating the impact of environmental agents and chemical products on thyroid function.

273 Elevated Ovarian Follicular Cell Apoptosis and Stress Protein Expression in Fish Exposed to Pulp Mill Effluents. Janz, D.M.*, Oklahoma State University, Stillwater, OK; McMaster, M.E., Munkittrick, K.R., Environment Canada, Burlington, ON; Van Der Kraak, G., University of Guelph, Guelph, ON. The objectives of this study were to determine the extent of apoptotic DNA fragmentation and expression of the 70 kDa stress protein (hsp70) in ovarian follicular cells of white sucker (*Catostomus commersoni*) exposed to pulp mill effluents. Fish were collected upstream and downstream of two mills in northern Ontario in order to compare potential effects of bleached Kraft (100% chlorine dioxide substitution) *vs.* thermomechanical effluent. Apoptosis was quantified using 3'-end labeling of ovarian follicular cell DNA with [^{32}P]dd-ATP. Ovarian hsp70 expression was determined using Western immunoblotting. Vitellogenic white sucker collected downstream of the bleached Kraft mill displayed a four-fold greater extent of ovarian apoptotic DNA fragmentation in comparison to fish collected upstream ($p<0.05$). The elevated apoptosis in exposed fish was associated with reduced gonadosomatic index (GSI; $p<0.001$) and increased ovarian hsp70 expression ($p<0.05$). There were no significant differences in plasma concentrations of testosterone or 17β-estradiol between female fish collected upstream and downstream of the bleached Kraft mill. In contrast to the Kraft mill, there were no differences in the extent of ovarian cell apoptosis, hsp70 expression or GSI in female fish collected upstream and downstream of the thermomechanical mill, although plasma testosterone and 17β-estradiol levels were reduced in fish collected downstream ($p<0.01$). These data suggest elevated ovarian cell apoptosis and hsp70 expression may be a result of exposure to bleached Kraft mill effluent components. Apoptotic cell death is the molecular mechanism responsible for ovarian follicular atresia and is regulated largely by hormonal factors; however these data and the results of a previous study (Janz *et al.* 1997, *Toxicol. Appl. Pharmacol.* 147, 391) indicate that alterations in circulating steroid hormones may not be associated with increased follicular atresia in fish exposed to pulp mill effluents.

274 Effects of Polluted Environment on Thyroidal Status of Mummichogs (Fundulus Heteroclitus). Zhou, T.*, Rutgers University, Newark, NJ; John-Alder, H.B., Rutgers University, New Brunswick, NJ; Weis, P., UMDNJ-NJ Medical School, Newark, NJ; Weis, J.S., Rutgers University, Newark, NJ. Previous studies have demonstrated that mummichogs (Fundulus heteroclitus) collected from Piles Creek (PC), New Jersey (a site polluted with heavy metals and organic materials) were sluggish and showed poorer prey capture and predator avoidance ability than those from a references site (Tuckerton-TK, New Jersey). In view of the considerable interest in "endocrine disruption" caused by environmental contaminants, we hypothesized that the behavioral dysfunction of the PC fish might be associated with thyroid impairment due to toxic exposure. In the preliminary study, we found that in comparison to TK, PC fish had larger thyroid follicles, greater follicle epithelial cell heights, and also contained significantly higher plasma thyroxine (T4) levels. PC fish exhibited a lower plasma T3/T4 ratio. However, there were no significant differences in levels of both plasma and tissue triiodothyronine (T3). There was also no difference in T4 and T3 levels between females and males. The differences in thyroid structure and function may be in part responsible for the behavioral differences between fish from the polluted PC and reference TK. In our ongoing experiments, TK fish are raised in simulated PC environment to determine if the polluted environment can alter thyroid function and spontaneous activity. In addition, PC fish are maintained in clean synthetic sea water to see if the thyroid alteration is recoverable.

275 Effects of Fenitrothion and Aldicarb Exposure on Cytochrome P450-catalyzed Estradiol Hydroxylase and Ethoxycoumarin O-deethylase in Channel Catfish. Perkins, E.J., and Schlenk, D. Environmental Toxicology Research Program/RIPS, Department of Pharmacology, School of Pharmacy, University of Mississippi, University, MS. In spite of the great amount of recent interest in endocrine effects in aquatic species, little effort has been made to determine the effects of environmental contaminants on the constitutive enzymes controlling sex steroid metabolism in fish. The objective of this project was to examine the effects of sub-lethal exposure to two representative agricultural insecticides, the carbamate aldicarb and the phosphorothionate fenitrothion, on constitutive cytochrome P450 (CYP) activity related to estradiol (E2) metabolism. Juvenile (1 year post-hatch, 172± 30 g) channel catfish (*Ictalurus punctatus*) were exposed to 0.5 ppm of each pesticide under static conditions for 24h (100% renewal at 12h). Hepatic microsomes were prepared and examined for CYP activities (Ethoxycoumarin-O-deethylase (ECOD) and estradiol hydroxylase (E2-OHase)). ECOD activity was determined using a microplate fluorescence assay. In addition to *in vivo* treatment effects on ECOD activity, *in vitro* inhibition by E2, fenitrothion, and aldicarb was assessed. Estradiol-hydroxylase activity was determined by tritium-release assay using 2,4,6,7-^{3}H-estradiol as substrate. CYP2-related isoform expression was determined by Western blotting with anti-CYP2M1 polyclonal antibody provided by Dr. D. Buhler. ECOD activity was significantly decreased (63% inhibition) by fenitrothion exposure; aldicarb had no significant effect. Estradiol and fenitrothion both were potent inhibitors of ECOD *in vitro*, with E2 displaying competitive type inhibition (k_i=3.9μM). Aldicarb and aldicarb sulfoxide displayed weak concentration-dependent inhibition of ECOD (28% and 29% respectively at 300μM). No effects on E2-OHase were elicited from *in vivo* pesticide treatment, but both compounds inhibited E2-OHase *in vitro.* Western blot analysis showed a significant decrease in a CYP2 isoform in the fenitrothion treated group and a significant correlation ($P < 0.05$; $R^2 = 0.471$) of this protein and ECOD activities among all treatment groups (n=9). These results suggest that exposure to some commonly used phosphorothionate insecticides has the potential to alter the normal estrogen biotransformation pathways in catfish.

276 Multi-Tiered Approach to Detecting PAH Effects on Grass Shrimp. Oberdörster, E.*, Martin, M., McLachlan, J.A., Tulane/Xavier Center for Bioenvironmental Research. New Orleans, LA. The steroid hormone ecdysone controls molting and differentiation in Arthropods. We used a three-tiered approach to investigate whether PAHs interact with the functional Ecdysone Receptor (EcR) *in vitro* and *in vivo*. We focused on four PAHs: pyrene, benzo[a]pyrene, benzo[b]fluoranthene, and chrysene, which are the most prevalent PAHs in Bayou Trepagnier, which has been heavily contaminated by the petroleum industry. A Chinese Hamster Ovary (CHO) cell line, which stably expresses the EcR/RXR complex, was used to determine reporter gene activation by PAHs either agonistically or antagonistically, and for binding assays using [^{3}H]-Ponasterone A. An ecdysone-responsive cell line (C18+) was used to determine whether PAHs interacted with the endogenous EcR/USP. Finally, grass shrimp (*Palaeomonetes* spp.) were used to assay for changes in molting after PAH exposure *in vivo*. In the reporter gene assay, none of the PAHs had any agonistic activity, but were each able to enhance reporter gene expression in the presence of the potent EcR agonist, muristerone A. In the C18+ cells, PAHs alone had no significant effect, while the PAHs in conjunction with muristerone A tended to enhance the cellular response (only B[b]F was significant). We concluded that PAHs were able to modulate ecdysone-dependent gene expression. However, the question of bioavailability, metabolism, and P450 induction were not taken into consideration in these *in vitro* studies. Ablated grass shrimp were exposd to chrysene in 48 hour static renewal. Shrimp molted approximately one day earlier after exposure to 100 nM chyrsene than the vehicle controls. Since chrysene was able to enhance reporter gene activation and the C18+ cellular response in the presence of the EcR agonist, the accelerated molting correlates well with the *in vitro* assays.

277 The Ability of Mammalian Antiandrogens to Bind to Androgen Receptors in the Brain of Rainbow Trout (*Oncorhynchus mykiss).* Wells, K.*, Van Der Kraak, G., University of Guelph, Guelph, Ontario. There is uncertainty whether compounds which bind to hormone receptors in mammalian species exert similar modes of action in non-mammalian species. In this study, we tested compounds that have been demonstrated to bind to androgen receptors in rats, to see if they bind with a similar affinity to an androgen receptor in fish. Cytosolic androgen receptors isolated from the brains of rainbow trout *(Oncorhynchus mykiss)* displayed high affinity for testosterone (Kd~1.3-2.0nM) and were extremely abundant

(~1230-1650fmol/mg protein). 5α-Dihydrotestosterone effectively competed for binding to the androgen receptor, in contrast to 11-ketotestosterone which bound poorly. Compounds which bind to the mammalian androgen receptor, 17β-estradiol, flutamide and cyproterone acetate, did not compete for binding to the androgen receptor in the rainbow trout brain. In addition, p,p'-DDE, which is a metabolite of DDT, and M1 and M2, which are metabolites of the pesticide vinclozolin, bind to the mammalian androgen receptor, but not to the rainbow trout brain androgen receptor. These results demonstrate marked differences between teleost and mammalian androgen receptors and call into question the applicability of making cross-class comparisons when evaluating suspected endocrine disrupting chemicals.

278 Time and Dose Dependence of Vitellogenin Appearance in Male Trout Plasma. Schultz, I.R. *, Battelle PNNL, Richland WA, Drum, A.S. Battelle MSL, Sequim WA and G A Orner, Battelle PNNL, Richland, WA. A commonly used endpoint in bioassays of the estrogenicity of chemicals is the induction of the female egg protein vitellogenin (Vg) in male fish. Typically, these studies administer the xenoestrogen via water exposure or ip injection and sacrifice fish at one or a few time points to monitor plasma levels of Vg. However, induction of Vg is a dynamic process and the lack of detailed knowledge regarding induction and elimination rates of Vg in fish can make it difficult to assess the degree of induction following exposure to a xenobiotic. In this study, we administered ethynylestradiol (EE2) at 0.001, 0.1, 1.0 and 10.0 mg/kg, p,p-DDT and DDE (1.0 mg/kg) to male rainbow trout via a dorsal aortic cannula which allowed repetitive blood sampling from individual fish for up to 48 days after injection. The plasma concentrations of Vg were quantified using ELISA and concentrations of EE2 & DDT & DDE determined by GC-MS / ECD. The pattern of induction of Vg was similar for all doses of ethynylestradiol, with a slight increase from basal levels of Vg (< 0.2 mg/ml) during the initial 24 hrs, then increasing sharply to maximum levels by days 8-12 (Cmax for 0.1-10 mg doses = 12, 25 and 42 mg/ml Vg) and then rapidly declining to near basal levels by days 16-24. Trout administered DDT or DDE showed no induction of Vg despite plasma concentrations of these chemicals comparable to the 10 mg/ kg ethynylestradiol treated fish. In a separate experiment, the plasma t1/2 of Vg was found to be 4.8 days in trout directly injected with purified Vg obtained from donor fish.

279 Synthetic and Natural Hormones in Wastewater Effluent: a Connection Between Pharmaceutically Active Compounds and Endocrine Disruption? Huang, C.H.; Chien, M.H.; Sedlak, D.L.*, University of California, Berkeley, CA. Predictions based upon prescription records and concentrations measured in urine indicate that synthetic and natural hormones (*e.g.*, ethinyl estradiol and 17b-estradiol) could be present in wastewater at concentrations as high as several parts-per-trillion. Comparison of these data with results of laboratory studies suggest that natural and synthetic hormones could be responsible for endocrine disruption in fish living in effluent-dominated waters; however, the actual concentrations of hormones discharged by wastewater treatment plants is unknown. To assess the fate of hormones during wastewater treatment, we measured concentrations of hormones at wastewater treatment plants equipped with a range of treatment technologies. Quantitative analysis of hormones, with detection limits below 1 ng/L, was performed by analyzing purified extracts with enzyme-linked immunosorbent assay (ELISA). Confirmatory analysis was performed using gas chromatography of fluorinated derivatives. Our measurements indicate that inactive glucuronide-conjugated hormones are converted into active hormones prior to wastewater discharge. Our measurements also indicate that little removal of hormones occurs in most conventional wastewater treatment plants. In all cases, concentrations of ethinyl estradiol and 17b-estradiol in effluent discharged by conventional treatment plants were significantly higher than those that reportedly cause endocrine disruption. Measurements made after advanced treatment processes (*e.g.,* microfiltration and reverse osmosis) suggest that hormones can be removed from wastewater effluent with existing technologies.

280 Input / Output Balance of Estrogenic Active Substances in a Major Municipal Sewage Plant in South Germany. Körner, W.*, Bolz, U., University of Tübingen, Tübingen, Germany, Hanf, V., University Women's Hospital, Ulm, Germany, Hagenmaier, H., University of Tübingen, Tübingen, Germany. Levels of estrogenic active substances were determined in parallel in the raw and treated sewage of the municipal sewage plant of Ulm, Germany, to evaluate the persistence of these compounds during modern wastewater treatment and to determine the magnitude of their release into rivers. Time proportional samples of sewage influent and effluent were taken automatically over 24 hours in the sewage plant. According to the average residence time of the sewage, effluent samples were taken 8 h later. Solid phase extraction of 1 L sample was performed on a polystyrene copolymer phase followed by elution with acetone. An aliquot of 10 % was analyzed for known phenolic xenoestrogens by GC/MS. In the remaining extract total concentration of estrogenic active compounds regardless their chemical structure was assessed by measuring the proliferative effect on estrogen receptor positive human MCF-7 breast cancer cells (E-screen assay) and comparing the EC_{50} values with those of the positive control 17ß-estradiol. Among nine analyzed phenols 4-t-octylphenol (OP), 4-nonylphenol (NP), Bisphenol-A (BPA), p-chloro-o-cresol, and 2-hydroxybiphenyl were found in the sewage influent in the upper ng/L range. While virtually the same levels of OP and NP were present in the effluent, BPA levels were reduced by 72 % and the latter two chemicals were eliminated quantitatively. Expressed in 17ß-estradiol equivalent concentrations (EE) total estrogenic activity was 38 ng EE/L in the influent and 4.3 ng EE/L in the effluent resulting an overall elimination rate of estrogenic compounds of 89 %. Thus, the estrogenic activity in the effluent was well within the range we recently found in four other sewage plant effluents in Germany, levels which are known to induce estrogenic effects in fish. As the detected levels of phenols only partly explain total estrogenicity in the effluent further analytical studies are necessary to identify all estrogenic substances.

281 Environmental Significance of DOM-Photogenerated Perhydroxyl Radical. Burns, S.E.*; Basford, T.M., Western Michigan University, Kalamazoo, MI; Hassett, J.P., SUNY-CESF, Syracuse, NY. Hydrogen peroxide in natural waters is produced through a sequence of reactions initiated when photoexcited DOM reduces oxygen to form superoxide anion. At environmental pHs, dismutation of superoxide to produce hydrogen peroxide will occur predominantly via the reaction of superoxide with its acid, perhydroxyl radical. Quantum yields and production rates have been thoroughly mapped out for the reaction sequence producing hydrogen peroxide from DOM via superoxide. However, the environmental significance of perhydroxyl radical remains largely unknown. We now report the inadvertent discovery of an environmental reaction mediated by perhydroxyl radical: the DOM-mediated transformation of quadricyclane. Based on that transformation, production rates for perhydroxyl radical are surprisingly high.

282 Production of Nitrite from the Photodegradation of Humic Substances in Natural Waters and Rainwater. Kieber, R.J.* , Li, A., Seaton, P.J., Willey, J.D. University of North Carolina at Wilmington, Wilmington, NC Significant concentrations of nitrite NO_2^- were produced from the photodegradation of humic substances (HS) isolated from a variety of natural waters in coastal North Carolina. Nitrite concentrations were 40 to 118 % higher after light exposure relative to initial levels, while no statistical differences were observed in dark controls before and after irradiation. The amount of nitrite produced upon irradiation was positively correlated to the concentration of HS added ($r = 0.97$, $p<0.001$). Production was also related to the length of time samples were photolyzed, with 80% of the increase occurring within the first two hours of the six hour exposure. The average production rate, normalized to both humic concentration and time of light exposure, for all humics studied ranged from 7.4 to 18.5 [nM hr^{-1} mg^{-1}] x 10^3. When natural waters were irradiated changes in nitrite were proportional to both initial nitrite and humic concentrations. Natural waters containing low initial NO_2^- (80 nM) gave a net NO_2^- increase of 4 nM after photolysis, while water with higher NO_2^- initially (800 nM) showed a net decrease of 132 nM after photolysis. The net loss or gain in NO_2^- upon photolysis can be understood as the competition between direct NO_2^- photolysis and production via HS photodegradation. Preliminary studies of the photochemical degradation of rainwater humic DOM are also presented. The photochemical release of biologically and chemically labile nitrite from biologically refractory humic substances has significant implications with respect to nitrite biogeochemistry and N cycling in natural and atmospheric waters. These results also suggest humic bound N is more biogeochemically labile than previously thought.

283 The Effect of Fe on the Rates of Cdom Photobleaching. Voelker, B.M.* and Ho, S.P., Massachusetts Institute of Technology, Cambridge, MA 02139. The goal of this study was to examine whether Fe can accelerate the rate of photobleaching of CDOM and, if so, what mechanisms are responsible. In a series of laboratory experiments, the rate of photobleaching of a humic acid was examined at different wavelengths as a function of pH and in the presence and absence of added Fe (10 mM). The humic acid had been acid-cleaned to remove most of the Fe initially present. Samples were irradiated using a medium-pressure Hg lamp equipped with a pyrex sleeve to block light of wavelengths less than 300 nm. Photobleaching was measured spectrophotometrically as loss of absorbance; measured absorbance values were corrected for the absorbance due to Fe in the solutions. In the absence of added Fe, photobleaching rates were not strongly affected by pH and absorbance at longer wavelengths was bleached more rapidly than absorbance at shorter wavelengths. The added Fe was found to have the greatest effect on photobleaching rates at acidic pH values. At a pH of 3, absorbance at 450 nm was bleached at twice the rate in the presence of Fe than in its absence. At 350 nm, acceleration by a factor of three was observed. At 250 nm, no bleaching was observed in the absence of added Fe while significant bleaching was found in its presence. Similar effects were observed at pH 4 but not at pH 7.5. Experiments are now in progress to examine the relative roles of HO radicals produced by Fenton's reaction (oxidation of Fe(II) by hydrogen peroxide, both photo-produced) and ligand-to-metal charge transfer reactions of Fe(III)-humate complexes in the observed photobleaching.

284 Atmospheric water: Transformation of Ozone into OH radicals by Sensitzed Photoreactions. Hoigné, J., Jans U., Swiss Federal Institute for Environmental Science and Technology (EAWAG), CH-8600 Dübendorf, Switzerland. The direct day-light photolysis of tropospheric ozone (O_3) in atmospheric waters proceeds only slowly. Rather, dissolved chromophoric compounds, such as ubiquitous iron(III)-oxalato complexes, act as primary photoreactants. Upon their fast photolysis in daylight, these produce hydroperoxyl radicals ($HO_2^{\bullet}$). Its dissociated form, the peroxide anion ($O_2^{\bullet -}$) transforms O_3 that is steadily supplied from the gas-phase and assumes about 1 nM. But already in presence of traces of dissolved copper at >1 nM, $O_2^{\bullet -}$ rather reduces copper(II) to copper(I) which then indirectly converts O_3 to $OH^{\bullet}$. Formate and formaldehyde at ubiquitous concentrations convert a fraction of the non selective $OH^{\bullet}$ to highly selective $HO_2^{\bullet}$ and $O_2^{\bullet -}$ (i.e. $HO_2^{\bullet}/O_2^{\bullet -}$). By passing the copper loop, this reacts with further O_3 to produce further $OH^{\bullet}$. However, ubiquitous acetate and some other types of solutes scavenge $OH^{\bullet}$ without recycling $HO_2^{\bullet}/O_2^{\bullet -}$. In proportion to their concentration relative to formate and formaldehyde, such solutes, therefore, limit the kinetic length of the O_3 converting chain reaction. Within cloud droplets these O_3 promoted aqueous-phase photoradical-type chain-reactions significantly increases the in-droplet flux of $OH^{\bullet}$ and $HO_2^{\bullet}$ radical. O_3 thereby acts as a chemical amplifier . Such reactions enhance aqueous-phase oxidation processes. Many of the applied reaction rate-constants have been measured earlier by pulse radiolytic methods (combined with kinetic spectroscopy) to produce $HO_2^{\bullet}/O_2^{\bullet -}$ .and to directly observe its reaction products. To better simulate the day-time cloud droplet conditions, including the slow radical flux, an *in vitro* production of $HO_2^{\bullet}/O_2^{\bullet -}$ by photolysing iron(III)-oxalate was, however, a more convenient tool. *This study is complementary to the EUROTRAC-HALIPP project.*

285 Uv-induced Redox-cycling of Iron and Dnom Photooxidation in the Epilimnion of Swiss Lakes. Emmenegger, L., Sulzberger, B.*, Sigg, L., Kaiser, E., Swiss Federal Institute for Environmental Science and Technology (EAWAG), CH-8600 Dübendorf, Switzerland. Photooxidation of dissolved natural organic matter (DNOM) is now known to be a significant sink of biologically refractory DNOM in marine systems and probably in freshwater systems as well. In iron-rich surface waters, light-induced processes involving Fe(III) complexes may play a significant role in controlling the fate of refractory DNOM. Conversely, naturally occurring organic ligands determine to a large extent the kinetics of (photo)chemical Fe(III) reduction and of Fe(II) oxidation, and thus iron speciation, which controls its bioavailability. We have conducted laboratory studies on the kinetics of Fe(II) oxidation in natural water samples from a eutrophic Swiss lake (Lake Greifen). Fe(II) concentrations were followed using an automated flow injection analysis system employing luminol-based chemiluminescence detection of Fe(II) to concentrations below 1 nM. The rates observed in raw, filtered and UV-treated samples indicate the existence of organic Fe(II) complexes which are rapidly oxidized by oxygen. We have also studied the light-induced reduction of naturally occurring Fe(III) in raw samples from Lake Greifen. Field studies on the diurnal Fe(II) cycling were undertaken in two Swiss lakes (Lake Greifen and Lake Tannen) that represent different conditions with respect to iron concentrations (due to different geochemical background), DNOM content, and light intensity (due to different altitudes). Using the data obtained from the laboratory and field studies, insight into the importance of the light-induced iron cycling on the fate of refractory DNOM in these lakes can be gained.

286 Photochemical Oxygen Consumption by Colored Dissolved Organic Matter in Marine Waters. Zafiriou. O. C.*, Woods Hole Oceanographic Institution., Woods Hole, MA; Andrews, S. S. Department of Chemistry, Stanford University, Palo Alto, CA; Caron, S., Woods Hole Oceanographic Institution., Woods Hole, MA. Solar photolysis of humic substances produces numerous chemical changes, such as decreases in fluorescence and absorption; formation of HOOH, CO, CO2, and numerous other trace products; and oxygen consumption. As multiple reactions are occurring in heterogeneous, ill-defined pools, the total change is unquantifiable. However, total oxidative equivalents of change are expected to be well quantified by net oxygen consumption, since few other oxidants are kinetically or thermodynamically competitive with molecular oxygen in natural waters. We measured photochemical oxygen consumption quantum yields (Φ, absorption-normalized) in several air-saturated coastal marine samples (0.2 micron filtered) as functions of absorbed dose (310 nm), wavelength, light intensity, and dilution with highly transparent, oligotrophic water. As the O2 uptake signals were very small, pulsed-mode dissolved oxygen electrodes (Endeco®) were used in conjunction with a dark-light-dark protocol to provide electrode drift corrections. Photochemical oxygen consumption was measurable in all three waters studied. It was wavelength-dependent, Φ values falling off approximately linearly with increasing wavelength in the UV region, 300-400 nm. A simple model fit to the time (dose) courses of oxygen consumption and photobleaching of absorbance required three components: two small, highly absorbing, high-Φ pools, and a much larger slower-reacting pool. Calculations of global and regional coastal photooxidation rates based on these data (with or without extrapolation into the >400 nm region) combined with other conservative assumptions suggest that these rates are significant, even at high latitudes. Factors controlling the environmental partitioning of partially photolyzed humic-like substances between biological oxidation and photo-oxidative transformations remain unknown.

287 Dissolved Inorganic Carbon Photoproduction from Dissolved Organic Matter and Its Fractions. Bourbonniere, R.A.*, Edmondson, K., Dunnett, F. and Street, A., Environment Canada, Burlington, ON, Canada. Dissolved organic matter (DOM) and its sub-fractions, isolated by XAD-8 and XAD-4 resins, are exposed to natural sunlight and artificial light sources. The photoproduction of dissolved inorganic carbon (DIC) from DOM is measured for a suite of waters from various Canadian lakes and streams covering a wide variety of DOM concentrations. As well humic acid and resin-isolated fractions of fulvic acid from these waters are studied for DIC photoproduction. Results from several of the waters suggests that differences in character (i.e. fraction distribution) have an impact on the amount of DIC that can be produced photochemically under laboratory conditions. Varying environmental and experimental conditions can lead to the development of action spectra for DIC photoproduction, an aid to modelling the impact of UV radiation on natural DOM.

288 The Pathway of Photo-degradation (Bleaching) of Laurentian Fulvic Acid. Langford, C.H. and Bruccoleri, A. Chemistry, University of Calgary, Calgary, AB. The photo-degradation and bleaching of Laurentian Fulvic acid has been monitored by absorbance changes, fluorescence changes, and solid state ramp-CP 13-C NMR. Parallel processes in model systems have been examined. We propose a quinone based mechanism which is consistent with the overall results seen and the data we have previously accumulated on the primary quantum yields for solvated electrons, triplets, and fluorescence for this sample.

289 Comparing the Photo-oxidation of DOM from Diverse Environments. Miller, W.L.*, Johannessen, S.C., Moore, R.J., and Scully, N.M., Dalhousie University, Halifax, NS, Canada. Each year more data accumulates on the direct photochemical oxidation of DOM in aquatic systems. Based on recent publications, there is a growing perception that photo-mediated loss of DOC directly to DIC may be a critical process in balancing aquatic organic carbon cycles. Most data, however, has been generated in lakes and/or nearshore coastal waters. To expand this view, we have performed photochemical irradiations on water from these environments as well as from the Gulf Stream, the Bering Sea, upper Baffin Bay and Kane Basin. A solar simulation system with a sequential series of optical longpass cutoff filters through the UV provided action spectra for DIC photoproduction in each of these environments. This presentation will provide a synthesis of these projects as they relate to the role of photochemical oxidation in organic carbon cycles. Preliminary indications are that the potential for marine DOM to be directly oxidized by sunlight to DIC is very much lower than that of terrestrially derived DOM. Consequently, the significance of direct photochemical loss of DOC to DIC may be much more important in evaluating the fate of terrestrial DOM in the ocean than as a loss term for marine derived DOM.

290 Changes in Functionality of Aquatic DOM Upon UV Irradiation. Thorn, K.A., USGS, Arvada, CO. A knowledge of how the structural characteristics of the various biologically refractory DOM fractions relate to the quantity and identity of low molecular weight carbon and nitrogen compounds produced upon UV irradiation is desirable for a comprehensive understanding of aquatic biogeochemistry. Liquid phase C-13 NMR analyses have been performed on aquatic fulvic, humic, and hydrophilic acids photobleached in increments of 25% loss of initial absorbance at 465 nm. Successive losses of carboxylic acid, ketone and quinone, aromatic, secondary alcohol and carbohydrate carbons were observed. The end products were decarboxylated, hydrophobic, predominantly aliphatic structures that were resistant to further degradation. One of the implications from these results is that, because aliphatic carbon structures are resistant to UV irradiation, for any suite of DOM fractions, the mass of carbon that can be released photochemically is related to the concentrations of carboxylic acid, aromatic, secondary alcohol, and carbohydrate carbons. Solid state N-15 NMR spectra of fulvic, humic, and hydrophilic acids from the Okeefenokee Swamp and Hellerudmyra Pond revealed the presence of naturally abundant heterocyclic and peptide nitrogens. The potential of N-15 NMR for studying the photochemical release of nitrogen from humic substances will be reviewed.

291 Seasonal Patterns in Pcb Flux from the Sediments of Rivers: a Mechanistic Evaluation. J. Rhea, J. Connolly, K. Russell, J. Quadrini, Quantitative Environmental Analysis, LLC, and J. Smith, Aluminum Company of America. River systems in the northeastern U.S., including the Grasse River, NY, exhibit seasonal patterns in PCB exchange between contaminated surface sediments and overlying waters. In these systems, the greatest PCB flux appears during late spring/summer low flow conditions and the composition of PCB loadings is consistent with a sediment pore water exchange mechanism. Physicochemical, biological, and hydrodynamic factors were explored as possible causes for the observed seasonality in loading. Temperature effects on molecular diffusion and PCB partitioning between sediment particles and pore water appear to account for a large portion of the seasonality. However, additional mechanisms, possible related to spring high-flow event induced replenishment of surface sediment PCB concentrations or the emergence of aquatic macroinvertabrates may be partially responsible for the observed temporal patterns in sediment pore water PCB loadings. Understanding the mechanisms contributing to these seasonal patterns in sediment-water exchange are important because of their potential impact on surface water concentrations which determine aquatic biota exposure concentrations and downstream PCB transport.

292 Triphasic Desorption of Highly Resistant Chlorobenzenes, PCBs and PAHs in Field Contaminated Sediment. ten Hulscher, Th.E.M., Vrind, B.A., van den Heuvel, H., van der Velde, L.E., van Noort, P.C.M., Institute for Inland Water Management and Waste Water Treatment (RIZA), Lelystad, the Netherlands, Beurskens, J.E.M., Present address: Water Board Maaskant, Oss, the Netherlands, Govers, H.A.J., University of Amsterdam, Amsterdam, the Netherlands. Desorption kinetics and partitioning of chlorobenzenes, polychlorinated biphenyls and polycyclic aromatic hydrocarbons in long term field contaminated sediment cores and top later sediment were measured by gas-purging. Desorption from sediment was deduced to be triphasic: fast, slowly and very slowly desorbing fractions were distinguished. In both the sediment core and the top layer sediment no detectable fast fractions were present for all the compounds studied. These fractions were estimated from the desorption curves. This observation coincided with very high *in-situ* distribution coefficients for several PCBs and PAHs: 10-1000 times higher than literature values for short contact time experiments. For all sediment samples slowly desorbing fractions (rate constants (3-8)x10^{-3} h^{-1}) and very slowly desorbing fractions (rate constants (0.16-0.5)x10^{-3} h^{-1}) were determined. In some cases only the very slowly desorbing fraction was detectable. Desorption from field contaminated sediments with extended contact times may not be readily estimated from laboratory experiments in which contaminants have contact times with the sediment in the order of weeks.

293 The Contribution of Oxypahs to Total Pah Loading in Contaminated Sediments and Water-borne Particulate. McConkey, B.J., Huang, X.-D., Dixon, D.G., Greenberg, B.M. University of Waterloo, Waterloo, ON. On exposure to light many PAHs undergo oxidation reactions, forming products such as quinones and hydroxyquinones (oxyPAHs). PAH photoreactions can occur in both the atmosphere and aquatic systems, with subsequent deposition in sediments or adsorption to suspended particulate. It has previously been shown that oxyPAHs are frequently more cytotoxic to invertebrates, plants and micro-organisms than the parent compounds. Thus, oxyPAHs can present an additional source of toxicity at PAH contaminated sites. Sediment samples were collected from several southern Ontario sites of known PAH contamination, plus a reference site with very low sediment PAH concentrations. Suspended particulate matter was also collected from the water column at several sites. Sediment components were isolated from sediment samples using solvent extraction, and separated by a two-step fractionation procedure. An initial fractionation was conducted by normal phase HPLC, followed by a reverse phase HPLC separation and diode array detection. The 2-D separation procedure allowed for isolation and non-destructive identification of PAHs and oxyPAHs from within the sample matrix. The relative toxic contributions were compared for PAHs and oxyPAHs present within the sediment samples, and it was shown that oxyPAHs can be significant contributors to cytotoxicity in contaminated sediments.

294 Irreversible Adsorption Capacities and Irreversible Desorption of Chlorinated Hydrocarbons in Natural Sediments. Chen, W.*; Kan, A.T.; Tomson, M.B. Contaminant that resists desorption is a serious environmental concern due to their impact on fate, transport, exposure risk, regulation, and remediation. Previous work by the authors has shown that an irreversible compartment, with finite capacity, existed in sediments. The chemicals in the irreversible compartment desorb at a constant $\sim 10^{5.5}$ (Environ. Sci. Technol. 32: 892, 1998). The objective of this study is to compare the irreversible capacity and desorption of different compounds in different sediments. A new experimental protocol was developed to saturate sediments with chlorinated benzenes in a relatively short time. After adsorption, the readily desorbed fraction was stripped with Tenax polymer resin. Then, the desorption from the irreversible phase was studied by successive desorption with electrolyte solution. The time period for each desorption step varied from 0.5 to 40 days. The solution phase concentration does not varied with the desorption time. The typical solution phase concentrations were 0.6-0.9 μg/L for 1,2-dichlorobenzene, 3.5-4.8 μg/L for 1,4 dichlorobenzene, 0.8-1.5 μg/l for 1,2,4-trichlorobenzene. The corresponding solid phase concentrations were 3.2, 5.2, and 5.3 μg/g for these three compounds, respectively. The irreversible capacities were 3.2 -- 18 μg/g for 1,3-, 1,4-dichlorobenzenes, hexachlorobenzene, and hexachlorobutadiene in Lake Charles sediments. These data yield a of= $10^{5.0-5.7}$ for chemicals listed above, except for hexachlorobenzene. The corresponding value for hexachlorobenzene is $10^{6.7}$. Additional compounds and sediments are currently being tested. The relation between different chemical and sediment types and properties of resistant desorption will be discussed.

295 Metal Speciation at the Lake DePue Wildlife Management Area. Anderson, Paul R.*, Illinois Institute of Technology; Chicago, IL; Bunker, Grant B., Illinois Institute of Technology, Chicago, IL. Lake DePue is a backwater lake on the Illinois River. Between 1977 and 1982, approximately 450,000 cubic yards of sediment were removed and pumped from Lake DePue to a diked area within the adjacent DePue Wildlife Management Area (DWMA). These sediments contain elevated levels of a number of metals including cadmium, copper, lead, and zinc. As part of an ongoing study, we are using X-ray absorption spectroscopy (XAS) to describe metal speciation at the DWMA. XAS is an important tool in environmental chemistry because it describes the electronic environment of elements in both crystalline and poorly crystalline solids such as soil clay minerals, and it provides information on the coordination environment of soluble and surface sorbed species. We report results from XAS measurements taken at the Advanced Photon Source (APS) at Argonne National Laboratory. The APS has higher energy characteristics and a high photon flux that provide a good signal-to-noise ratio and make it possible to examine samples with relatively low metal concentrations. We compare sample measurements to absorption spectra from a basis set of reference compounds believed to represent species at the DWMA. The reference compounds include soluble metal-nitrate solutions, metals sorbed to clay or oxide minerals, and metal carbonate and sulfide precipitates. Most of our measurements to date focused on zinc. Our preliminary results suggest that zinc in oxidized soils is probably coordinated with oxide and clay minerals.

297 Reaction Pathway Analysis of Environmental Selenium in the Idaho Phosphate Resource Area. Bond. M., Steinhoff P., Geist, D. and Möller, G.*, University of Idaho, Moscow, ID. The observation of chronic selenium toxicosis in livestock and high environmental Se levels near the Idaho phosphate resource area led to an investigation of the source of the contamination. Water Se concentrations, sometimes exceeding 1 mg/L, have been observed in surface water resulting from leaching in the overburden waste rock dumps associated with phosphoria strip mining. Chemical and mineralogical studies of the waste rock, leachates and representative surface waters reveal a highly efficient mechanism for extraction of oxidized selenium as compared to heavy metals of interest. Microscopic and electron microprobe studies of polished thin sections indicate the presence of selenium containing pyrite in a siltstone fraction of the waste rock. A sequential extraction scheme on the waste rock soils indicate that selenite is the leachable moiety and that reduced selenium, as Se (0) or Se(-II), make up nearly 98% of the approximate 30 mg/kg total Se load. The role of organoselenium compounds in these organic rich soils is examined.

298 Estimation of Heavy Metal Bioavailability in Contaminated Soils by a Sequential Extraction Procedure. Basta, N.T. and R. Gradwohl, Oklahoma State University, Stillwater, OK. Heavy metal bioavailability of 15 contaminated soils ranging from 2.3 to 296 mg Cd kg^{-1}, 55 to 3180 mg Pb kg^{-1}, and from 432 to 23970 mg Zn kg^{-1} was estimated by the Potentially BioAvailable Sequential Extraction (PBASE). PBASE is a four-step sequential extraction; extraction 1 (E1) is 0.5 M $Ca(NO_3)_2$, E2 is 1.0 M NaOAc, E3 is 0.1 M Na_2EDTA, and E4 is 4 M HNO_3. Heavy metal bioavailability in two human exposure pathways, plant uptake and incidental ingestion, was determined from lettuce uptake and *in vitro* gastrointestinal studies. Lettuce uptake of Cd ($p<0.001$) and Zn ($p<0.05$) was predicted by the PBASE E1 extract. Gastric concentrations of Cd, Pb, and Zn were described ($p<0.001$) by the summation of all extracts (SE_{1-4}) or by total extractable metal. Both SE_{1-4} and total extractable metal accounted for 100% of gastric Cd and Zn but extracted twice the amount of gastric Pb. The PBASE E1 extraction accounted for 100% of intestinal phase Cd concentration. Intestinal Pb ($p<0.001$) was described but overestimated by E2 or SE_{1-2} extractable Pb. SE_{1-2} extractable Zn described ($p<0.001$) but slightly overestimated (14%) intestinal phase Zn. The PBASE method can be used to estimate Cd, Pb, and Zn bioavailability in smelter contaminated soils.

299 Nonequilibrium Sorption and Desorption Behavior of Phenanthrene in Rubbery and Glassy Organic Matter. LeBoeuf, E.J.*, Vanderbilt University, Nashville, TN; Weber, W.J. Jr., The University of Michigan, Ann Arbor, MI. Recent discovery of glass transitions within soil-derived organic matter suggests a direct link between the expanded and condensed domains of natural organic matter and the rubbery and glassy states of synthetic organic polymers, respectively. Nonequilibrium sorption studies of phenanthrene within natural and synthetic organic matrices with known glass transition temperatures have yielded evidence of concentration-dependent phase distribution relationships (PDRs) for natural and synthetic glassy sorbents, while rubbery matrices displayed linear PDRs over widely varying time intervals. This experimental evidence lends additional support to the hypothesis that diffusion within more diagenetically altered organic matter is likely nonFickian in nature (including both Case II and anomalous diffusion), and that mass transport within expanded, more rubber-like organic matter is likely attributable to simple Fickian diffusion. Sorption/desorption studies on the same sorbents have revealed direct evidence of significant desorption hysteresis for glassy matrices and a lack of desorption hysteresis for rubbery matrices. This additional evidence suggests that resistant sorption processes observed for organic compounds in natural particles are likely results of sorption and desorption processes associated with the more condensed, glass-like organic matter domain.

300 Fate of Spinosyns in Litter and Soils of a White Spruce Plantation in Central Ontario. Thompson, D.*; Buscarini T., Chartrand, D., Canadian Forest Service, Sault Ste. Marie ON.; Harris, B., DowElanco Canada, NewMarket ON. Spinosyns, a class of natural compounds produced by the actinomycete *Saccharopolyspora spinosa*, have significant insecticidal activity and resultant use-potential in Canadian forestry. Comparative field experiments were conducted under both full canopy and in a natural opening of a mature white spruce plantation of the Great Lakes-St. Lawrence forest region to assess terrestrial fate aspects associated with this potential use pattern. Residues of the parent active spinosyns (A and D), as well as their demethylated metabolites (DM-A and DM-D), were quantified via RP-HPLC analyses of litter and thatch of intact split-plots, as well as underlying soils of both intact and exposed split-plots. Results demonstrated that both principal active ingredients (spinosyn A and D), dissipated rapidly in the upper layers of the forest floor. Time to 50% dissipation ranged from 2.0 to 7.8 days depending upon matrix, experiment and split-plot condition. A general trend of more rapid dissipation in exposed soils, versus litter or thatch matrices, was observed. However, overlap in the 95% confidence limits for these endpoint estimates suggests that such differences are of academic rather than practical importance. By the end of the 39 day period of observation, dissipation of greater than 83% of the initial residues of parent spinosyns had occurred in all cases. Transient increases in the demethylated metabolites confirmed that the parent spinosyn molecules were being degraded *In-situ*. Analyses of underlying soil layers showed only sporadic low-level detections of parent spinosyns or their respective metabolites, confirming that none of the four spinosyn analytes were susceptible to leaching under these field conditions.

301 Indicators to Measure Eco-efficiency. Lehni M., World Business Council for Sustainable Development (WBCSD), Geneva. The concept of eco-efficiency, which the WBCSD has introduced and is steadily promoting, is being more and more recognized by various stakeholders and the entire business community as a successful way to achieve progress towards sustainability. To further develop the concept, the WBCSD is conducting a project to define a standardized methodology to measure and report eco-efficiency and thus the environmental performance relative to the economic performance of businesses. The project's deliverables, which are developed in expert workshops and stakeholder dialogues, are universal metrics principles, a harmonized set of crosscutting eco-efficiency indicators, guidelines to define additional sector-specific indicators, pilot applications by member companies, and useful application tools and training material to support the implementation of the metrics system. In a state-of-play report on eco-efficiency metrics WBCSD compiles -the current industry practice in environmental performance quantification, and analyzes respective stakeholder needs on metrics and reporting. –The report identifies a pressing need to develop universal environmental metrics. Ecometrics to be –broadly applied and accepted, need to –allow comparability and benchmarking over time and across products, –companies and industry sectors. –The basis for any sound metrics is to provide credible information which is timely, clear, verifiable, and comparable. WBCSD concludes the following environmental items to be quantified: energy, materials, water, waste and emission categories, such as global warming, ozone depletion, and acidification. As eco-efficiency includes both ecological and economic excellence, the respective performance metrics shall express the link to the bottom line. WBCSD has found that environmental measurements shall be normalized with respective economic terms. The environmental impact a particular business implies shall be related to the value added it is able to achieve.

302 Environmental Indicators and Measuring Environmental Performance - Perspective from a Consumer Goods Company. Shimp, R. J.*, Owens, J. W., The Procter & Gamble Company, Cincinnati, Ohio, USA. Environmental indicators are intended to translate complex environmental data into a form that can help businesses, governments, and public interest stakeholders evaluate environmental performance and analyze the condition of the environment. For business, environmental indicators have the following major uses: (1) Identifying opportunities for environmental improvements in products and processes; (2) Assessing the effectiveness of environmental management programs; (3) Establishing environmental goals; and (4) Communicating environmental progress. Environmental indicators may have similar applications in the context of public policy. For example, they can help guide government decision-making, aid efforts to establish environmental priorities, and help measure the effectiveness of policies and regulations. This paper describes a set of fundamental principles that can provide a useful framework for establishing environmental indicators, regardless of their application. Briefly, all indicators should be:

- Relevant and meaningful for protecting the environment and human health.
- Based on sound science.
- Strongly related to a defined environmental outcome.
- Clearly defined, and based on high quality, verifiable data.
- Flexible enough to accommodate the diversity of organizations.
- Transparent, understandable, and not potentially misleading.

Key issues associated with the development and use of indicators will also be discussed, especially in the context of P&G's global consumer products business.

303 Technical Evaluation Guidelines for Ecological Indicators. Jackson, L.E., U.S. EPA, Research Triangle Park, NC. EPA's Office of Research and Development (ORD) has prepared fifteen technical guidelines to evaluate the suitability of an ecological indicator for a particular monitoring program. The guidelines are organized within four recommended evaluation phases: conceptual relevance, feasibility of implementation, response variability, and interpretation and utility. ORD has adopted this evaluation process as iterative guidance for internal and affiliated researchers during the course of indicator development, and as a consistent framework for indicator review. The guidelines are described in Chapter One of the forthcoming EPA technical document, "Evaluation Guidelines for Ecological Indicators." Successive chapters illustrate application of the guidelines to diverse indicators in various stages of development. Example indicators range from direct chemical measures, such as dissolved oxygen concentration, to multi-metric biological indices like the fish index of biotic integrity. The purpose of these illustrations is to demonstrate the evaluation process using real data and working with the limitations of research in progress. Furthermore, these chapters demonstrate that an evaluation may emphasize individual guidelines differentially, depending on the type of indicator (ie. field versus remote, direct versus derived), and program design (ie. status versus trend, regional versus local). The evaluation process identifies weaknesses that may require further indicator research and modification. Alternatively, users may choose to adopt an indicator that exhibits selected strengths of particular importance to a program's objectives. The protocol in ORD is to demonstrate that an indicator performs satisfactorily in all phases before recommending an indicator for use. The EPA evaluation guidelines document represents a compilation and expansion of previous efforts, in particular, the initial guidance developed for EPA's Environmental Monitoring and Assessment Program (EMAP).

304 Development and Application of Chemical Exposure Criteria for Comparing Sites and Watersheds. Fulk, F.*, Lin, E., Subramanian, B., Schubauer-Berrigan, M., U.S. EPA, Cincinnati, OH; Williams, D., Sobran, Cincinnati, OH; Capuzzio, K., Counts, B., Altfater, D. OEPA, Columbus, OH; Cormier, S., U.S. EPA, Cincinnati, OH. This research investigated sampling and estimation approaches for measuring background levels of fish exposure to chemical contaminants. One research goal was to develop a method for deriving quantitative exposure criteria useful to compare a site or watershed to a reference condition. A second goal was to provide a foundation for managers in setting reasonable expectations for protecting stream quality. The prototype method used indicators of exposures to oil contamination and combustion by-products. The indicators were naphthalene and benzo[a]pyrene metabolites sampled from white suckers in the Eastern Cornbelt Plains Ecoregion of Ohio. Sites were selected by two sampling schemes. First, second and third order stream sites were selected by an intensified probability-based sampling design developed by the Environmental Monitoring and Assessment Program (EMAP). For larger streams, reference sites were selected by Ohio EPA ecologists. Development of the method for deriving exposure criteria included a comparison of parametrically and non-parametrically derived values for EMAP sites and OHIO EPÁ sites. Factors considered included stream order and site selection. The non-parametric approach, the 95th percentile derived from ranked data, was statistically robust. Exposure criteria were descriptive of exposures to fish and were approximately 25% higher for larger streams. An assessment of streams of concern in Ohio and a high quality watershed demonstrated that exposure criteria were practical for comparing and communicating exposures in streams. A poor quality stream segment demonstrated that exposure criteria were useful for monitoring change in exposure levels.

305 The Development of Environmental Indicators in Support of State Water Quality Management Objectives. Yoder, Chris O. Water quality management agencies have historically evaluated their success by using administrative and source-specific indicators such as permitting, compliance, enforcement actions, and gross pollutant loading reductions. Recently, U.S. EPA and others have undertaken efforts to promote the development and use environmental indicators. A hierarchy of administrative and environmental indicators was developed which promotes an organized and sequential use of both administrative and environmental indicators to evaluate the effectiveness of water quality management programs. Ohio EPA conducted a pilot study to evaluate this hierarchy and several of the national indicators developed by U.S. EPA. We were able to show associated relationships between some of these indicators and further demonstrate the importance of using each within their most appropriate roles as stressor, exposure, and response indicators. Our ability to test the national indicators was enhanced by an existing database and systematic monitoring and assessment program which spans more than twenty years throughout the state of Ohio. In one of the two case studies highlighted here we were able to associate the success of permitting, funding, and wastewater treatment plant (WWTP) construction to meet water quality-based objectives by linking these activities to reduced loadings of key pollutants, improved ambient water quality, and improvements in the biological response indicators. In the remaining case study, similar actions taken regarding permitting, funding, and WWTP construction also resulted in reduced pollutant loadings and improved ambient conditions, but virtually no change in the biological response indicators which continued to show severe degradation. The presence of other problems including contaminated sediments and recurring spills and releases essentially precluded any meaningful biological recovery. The use of a multiple lines of evidence approach in inferring causal associations between the available stress, exposure, and response indicators is highlighted by this case study. It also illustrates the importance of having a comprehensive monitoring and assessment effort on a watershed scale to ensure that major environmental problems are not overlooked or under-rated. The EPA indicators hierarchy promises to be an effective way to organize information about different indicators and provides a framework for the systematic evaluation of the results of State and Federal water quality management efforts.

306 Multi-layered Indicators using Geographical Information Systems for Consensus in Environmental Regulation. Louis, G.E., University of Virginia, Charlottesville, VA; Huang, S., University of Virginia, Charlottesville, VA; Plemmons, T.*, University of Virginia, Charlottesville, VA; Makowsky, M., University of Virginia, Charlottesville, VA. A risk communication tool was developed to facilitate participation in environmental regulation by stakeholders from the public, industry and government. The goal was to use information about environmental risk as the basis for consensus between stakeholders in environmental policy. The objective was to develop a set of indicators that would; graphically display expressed public concerns about environmental risk, superimpose correlation with industry presence onto this display, then add correlation with regional demographics to the display. Members of the University of Virginia and community groups in the City of Charlottesville were surveyed for public concerns about the health risks from environmental pollution caused by selected chemicals in the Toxics Release Inventory (TRI). An "expressed hazard ranking" was used to select the three most important contaminants in each medium. These constituted the environmental indicators for air, soil and water in the Charlottesville area. Data from the Virginia Department of Environmental Quality (DEQ) and EPA were used to generate air, soil and water layer maps in ArcView geographical information systems (GIS). Standard Industrial Classification (SIC) codes (the industry indicators) were used to add an industry layer to the Charlottesville map. Census data on race, income and education (the demographic indicators) formed demographic layers in the map. Charlottesville was then divided into four geographic quadrants of equal area. Spatial regression analysis across the four quadrants searched for relationships between the indicators: industry/environmental, industry/demographic and demographic/environmental. The results show correlation between industry/demographic and demographic/environmental indicators. Larger sample sizes from more cities are needed to validate these results and investigate the relationship between industrial and environmental indicators. The maps and results are readily displayed as Web pages. Thus, this tool provides accessible information on environmental risk to the primary bearers of environmental costs (the public), the primary beneficiaries from environmental loading (industry) and the government agencies responsible for arbitrating these costs and benefits in the interest of public health.

307 Sediment Contamination, Histopathology, Toxicity, and Community Composition in Chesapeake Bay Tributaries. Brown, S.*, Chesapeake Biological Laboratory, Solomons, MD; Dawson. C., Durell, E., McGinty, M., Maryland Department of Natural Resources, Annapolis, MD; Evans, J., Oxford Cooperative Laboratory, Oxford, MD; Ranasinghe, A., Versar, Inc., Columbia, MD. We collected and analyzed data from 13 muddy, low-mesohaline stations, including sites in the Patuxent, Magothy, Wicomico, and Chester rivers, to evaluate relationships between sediment contaminants and biological response variables. Significant negative correlations were found between chemical stressors and biological responses (e.g., chlorpyrifos vs. amphipod survival, metals vs. benthic species richness), and positive relationships were found between effects at different levels of organization (e.g., fish histopathology vs. amphipod/fish survival). Concentrations of metals exceeding ER-L thresholds were found in the Magothy (As, Cd, Cu, Hg), Patuxent (As, Cd, Ni, Zn), Wicomico (Cd, Ni) and Chester (As, Ni) rivers; metals exceeded ER-Ms only in the Magothy (Ni, Zn). Similar site-specific exceedences were found for organic contaminants (PAHs, PCBs, pesticides). Sediments from the Chester River elicited significant mortality among amphipods and fish, while sediments from Patuxent elicited significant mortality among amphipods (Eagle Harbor, Prison Point) or fish (Holland Bar, Buzzard Island, Sheridan Point), but not both. Sediments from the Magothy reduced fish survival and amphipod growth, and amphipod growth was reduced in Wicomico sediments. Histopathological analysis revealed that fish exposed to sediments from the Chester and Patuxent scored poorly , while those exposed to sediments from the Magothy and Wicomico scored average to good. Specific anomalies will not be discussed here. Benthic communities were severely degraded at all sites in the Magothy, and degraded at selected sites in the Patuxent and Wicomico rivers. Fish community composition was degraded at the Chester river site, but the benthos met restoration goals.

307a Scoring the Scorers: Evaluation Criteria for National and Global Environmental Indicator Systems. C.A. Pittinger, J.W. Owens, The Procter & Gamble Company, Cincinnati, Ohio. The sharp rise in the development of environmental indicator systems nationally and globally requires practitioners to make far-reaching policy decisions regarding the selection, application, interpretation and communication of environmental information. Indicators with wide-ranging objectives are currently being developed or applied by: the United Nations, the Organization of Economic Cooperation and Development, the European Union, the World Bank, the World Business Council for Sustainable Development, the U.S. Environmental Protection Agency, the Office of Science & Technology Policy, and others. *Condition indicators* have been proposed to characterize inherent, empirical attributes of the environment. In contrast, *performance indicators* are used to score functional aspects of environmental management systems, ranging across regulatory programs, corporate practices, and attributes of commercial products. Objectives are easily compromised when the goals of the program are not clearly articulated, when indicators are inconsistent with the program objective, or when distinct and unrelated endpoints are aggregated into overly simplistic scoring systems. This paper overviews the diversity of indicator systems and approaches currently under development, and contrasts their objectives, applications and approaches. Selected indicator systems will be evaluated on the basis of the following criteria: relevance to stated objectives; technical accuracy; transparency of methods; clarity in distinguishing correlation and causation among observed trends; validity of indicator aggregation methods; uses of complementary tools; and adherence to accepted risk communication principles.

308 Environmental Factors in Relation to Trace Elements in Streambed Sediment and Aquatic Biota of the Western Lake Michigan Drainages, 1992-1997. Scudder, B.C.*, Fitzpatrick, F.A., Sullivan, D.J., Rheaume, S.J., U.S. Geological Survey, Middleton, WI and Lansing, MI. Sampling was conducted in 1992, 1994, 1995, and 1997 to determine the occurrence of a broad suite of trace elements in streambed sediment and biota in selected streams in the Western Lake Michigan Drainages - a study unit of the National Water-Quality Assessment (NAWQA) Program of the U.S. Geological Survey. Sites were characterized with regard to dominant land use/land cover, bedrock type, and surficial deposit type in the basin using a geographic information system. Fine-grained sediment (< 63 micron) was collected from the upper 2 cm sediment layer at 31 sites; aquatic biota (primarily caddisfly larvae) was collected at 25 of the 31 sites. Some variability in trace elements was related to land use, however variations in bedrock type and (or) surficial deposit type complicate interpretation. Urban land use was the dominant factor correlating to high concentrations of Cd, Cu, Hg, and Pb in sediment. Igneous/metamorphic bedrock and forested land use was related to high concentrations of As in sediment and caddisfly larvae, whereas sandy/sand and gravel surficial deposit type and forested land use was related to high concentrations of Se in sediment. Significant positive relations were found between Se concentrations in sediment, caddisfly larvae, and sediment organic carbon. Land use, surficial deposits, and bedrock type were related to Ni and Zn concentrations in sediment; land use and surficial deposit type were related to Ni concentrations in caddisfly larvae. Cr concentrations in sediment and biota did not directly reflect any of these factors.

309 Geochemical Indicators of Intrinsic Remediation of Groundwater Contaminated with Volatile and Semi-volatile Organic Compounds. Pardieck, DL*[1], Carmichael, L.M.[2], Smith, T.S.[1], Sieber, E.[1]; 1, Ciba Specialty Chemicals, Toms River, NJ; 2, NC A&T State University, Greensboro, NC. Chlorinated organic compounds such as TCE and PCE are common pollutants present in the subsurface throughout the industrialized world. The purpose of this paper is to describe geochemical information that indicates whether intrinsic remediation is occurring in the subsurface of a site in southern New Jersey contaminated with a variety of volatile and semivolatile organic compounds. Intrinsic remediation was investigated in two sandy, low organic carbon aquifer units separated by a silty aquitard. Evidence of intrinsic remediation was based on groundwater sampling for a variety of chemical parameters (volatile and semivolatile organic compounds, electron acceptors and reduced species, methane, ethene, ethane, other anions and cations, CO_2, CH_4, pH, Eh, DO, and other parameters). Groundwater data indicated that subsurface regions with high concentrations of volatile organic compounds also had increased concentrations of CO_2, decreased pH/Eh and increased ferrous iron related to background groundwater. Methane production has also been observed in several monitor wells. Ethene was also observed, indicating complete reductive dehalogenation of chlorinated ethenes in the groundwater. These coinciding factors indicate that microorganisms are probably metabolizing the organic pollutants in the subsurface. In addition, the plumes at the site can be divided into three distinct groundwater zones, each of which show evidence of intrinsic remediation.

310 Relative Potency of Polycyclic Aromatic Hydrocarbons (PAH) for Inducing CYP1A1 in Juvenile Trout (*Oncorhynchus mykiss*). S.M. Billiard* and P.V. Hodson, Queen's University, Kingston, ON CANADA; N.C. Bols, Department of Biology, University of Waterloo, ON, CANADA. Retene (1-methyl-7-isopropyl phenanthrene) is an alkyl-substituted phenanthrene similar to PAHs found in petroleum and products of combustion. We have previously shown that chronic exposure of trout larvae to retene causes dioxin-like toxicity or blue sac disease. Symptoms included prolonged cytochrome P4501A1 (**CYP1A1**) induction, edema, hemorrhaging, and craniofacial abnormalities. Chronic exposure to retene resulted in mortality before swim up and recruitment failure. Results suggest that rapidly metabolized PAH share the same mechanism of toxicity as the more persistent and toxic chlorinated PAH (**CPAH**), i.e. oxidative stress due to prolonged CYP1A1 induction. If this model is correct then the chronic toxicity of PAH to larvae should increase with potency for MFO induction. The rank order of potency for CPAH in causing blue sac disease is the same as the relative potency of these congeners for inducing CYP1A1. Fish-specific toxic equivalency factors (**TEFs**), which relate the potency of individual congeners to dioxin, have been used to assess the risk of CPAH and of CPAH mixtures to fish early life stages. Because PAH are ubiquitous environmental contaminants the risk of toxicity from chronic exposure to PAH may be greater. CYP1A1 induction in juvenile trout was used to derive TEFs for 5 PAH chosen for their induction potency in a rainbow trout liver cell line (RTL-W1). The potency of PAH for causing induction was estimated from exposure-response curves as the median effective concentration (**EC_{50}**) and used to calculate TEFs. With the exception of phenanthrene, all PAH tested induced CYP1A1 activity in juvenile trout and were ranked as follows: benzo[k]fluoranthene > β-naphthaflavone > benzbfluorene > retene. These rankings agree with TEFs derived from RTL-W1 cell lines and are currently being used to determine if short-term CYP1A1 induction in juvenile trout can predict chronic toxicity to larvae of PAH and PAH mixtures. This may provide the tools for rapidly assessing the risk of PAH from any source causing recruitment failure of fish.

311 Specific Accumulation of Butyltins and Total Tin in Marine Mammals. T. H. L. Le*, N. Nakatani, S. Takahashi and S. Tanabe, Ehime University, Matsuyama, Ehime, Japan; K. Saeki, Oita University, Oita, Japan; N. Miyazaki, The Univ. of Tokyo, Iwate, Japan and Y. Fujise, Inst. of Cet. Res., Tokyo, Japan. Concentrations of total tin (Sn: organic + inorganic) and total butyltin compounds (BT: MBT+DBT+TBT) were determined in various tissues and organs of marine mammals such as cetaceans and pinnipeds collected from Japanese coastal waters. Additionally, some marine mammal species from Hong Kong, Philippines, India, the Black Sea, the Pacific and Antarctic Oceans were also subjected to those analyses for comparison. In most species analyzed, ?BT concentrations were found to be comparable with those of Sn. Concentrations of Sn as well as BT were highest in liver among organs and greater in coastal species than in offshore species. Hepatic concentrations of Sn increased with an increase in BT residues ($r = 0.95$, $p < 0.001$) and percentages of total butyltins in total tin (BT/Sn) were found considerably high in coastal cetaceans. These results indicated that the hepatic tin in the coastal cetaceans predominantly exists in the organic form as butyltin compounds, implying that Sn residues in marine mammals reflect mostly input from anthropogenic sources. Concentrations of Sn and BT found in the liver of pinnipeds were significantly lower than those in cetaceans, suggesting their excretion through metabolic degradation and shedding in pinnipeds.

312 Contamination by Persistent Organochlorines in Cetaceans from Hong Kong Coastal Waters. Tu B.M.*, Watanabe, M., Nakata, H., Tanabe, S. (Department of Environment Conservation, Ehime University, Japan) and Jefferson, T. (Ocean Park, Hong Kong. Blubbers samples of 2 cetacean species : Indo-Pacific Humpback dolphin (*Sousa chinensis*) (n = 11) and finless porpoise (*Neophocaena phocaenoides*) (n = 9) collected from Hong Kong coastal waters were analysed for persistent organochlorines. Mean concentrations in the blubbers of DDTs were ranked first (70 μg/g wet wt.), followed by PCBs (37 μg/g wet wt.), HCHs (0.84 μ/g wet wt), chlordane compounds (0.51 μg/g wet wt.) and HCB (0.08 μg/g wet wt.). It has been seemingly suggested that the present cetacean species may pottentially be faced to high risk due to the elevated level of DDTs and PCBs. Futhermore, these relatively high concentrations of PCBs and DDTs can be attributed to the continuous environmental input of these compounds in the Far East region including Hong Kong. While attempting to understand the contamination status in Hong Kong cetaceans, *tris*(4-chlorophenyl) methane (TCPMe) and *tris*(4-chlorophenyl) methanol (TCPMeOH), which are among the most recently-identified organochlorine microcontaminants, were also detected with the highest concentrations of 240 ng/g and 270 ng/g (wet wt. basis), respectively. The concentration of TCPMe relative to TCPMeOH in cetaceans from Hong Kong coastal waters was significantly higher than those found in various seals collected from other parts of the world, suggesting the different metabolic capacity of these two compounds between seals and cetaceans. Correlation between the concentrations of *tris*-chlorophenyl compounds with other persistent organochlorines such as HCHs, CHLs, DDTs and PCBs was significant, which may suggest their accumulation nature similar to these organochlorines.

313 Trends in organochlorine concentrations in grey seals (*Halichoerus grypus*) from Sable is., NS, 1974-1994. Addison R.F.* DFO Institute of Ocean Sciences, Sidney, BC Canada; Stobo, W.T., DFO Bedford Institute of Oceanography, Dartmouth, NS, Canada. Reproducing female grey seals (*Halichoerus grypus*) have been sampled from Sable Is., NS, for analysis of organochlorines (OC) in blubber, at intervals during the last 25 years. Between 1974 and 1994, concentrations of the DDT-group of insecticides declined about 20-fold, from a mean of approximately 12 μg/g lipid to approximately 0.5 μg/g lipid. The fraction of the DDT-group represented by *p,p'*-DDE increased from about 60% to about 90% over this interval. Concentrations of polychlorinated biphenyls (PCB) also declined, though more slowly, from a mean of approximately 20 μg/g lipid (expressed as Aroclor 1254) to about 7 μg/g lipid in 1994. No clear trends were detectable in congener distribution between the mid-1980's (when individual congeners were first analysed) and 1994. Between 1984 and 1994, α-hexachlorocyclohexane concentrations declined by about half, but those of hexachlorobenzene and of oxychlordane were unchanged. Throughout the period 1974-1994, the Sable Is. grey seal population grew significantly, and the total burden of DDT-group residues in the population may not have declined as dramatically as is suggested by the analyses of the individuals sampled.

314 Patterns and Risk of Organochlorine Bioaccumulation in a Northwest Atlantic Food Web. Weisbrod, A.V.*, Shea, D., LeBlanc, G.A., North Carolina State University, Raleigh, NC; Moore, M.J., Stegeman, J.J. Woods Hole Oceanographic Institution, Woods Hole, MA. The crash of several New England fisheries, the 1988 bottlenose dolphin mass mortality along the mid-Atlantic seaboard, and the lack of recovery of multiple whale populations raise concern for the integrity of our Eastern U.S. marine environment. Over fifty organochlorines were measured via dual column GC/ECD in samples from seven trophic levels of the food web shared by long-finned pilot whales, white-sided dolphins, and endangered right whales. Using visual plots and multivariate statistical techniques, primary factors influencing trophic level bioaccumulation and sample variation were identified. We find that the bioaccumulation of 4,4'-DDE, several chlordanes and PCB congeners is substantial (ppb to ppm) in the stranded odontocetes and squid. Fish destined for human market and right whale biopsies were 1-2 orders of magnitude less. This trend is consistent with the few reports of other NW Atlantic cetaceans. Plots of PCB metabolism groups provide insight into trophic transfer dynamics and aid in the identification of likely whale food sources. Principle component analysis was used to identify several factors that appear to govern bioaccumulation patterns: exposure to different food sources, season of sample collection, PCB congener metabolism, tissue type, gender and maturity. In addition, dolphin and pilot whale viscera contain ample levels of specific compounds shown to alter endocrine function. We propose a mechanism-based scheme for extrapolation of potential endocrine disruption in these protected species based on receptor homology, comparable biochemical pathways, and serum hormone levels.

315 Global Transport Processes and the Trophic Transfer of Xenobiotics: Implications for Future Food-Chain Transfer to Humans and Marine Mammals. Shea, D.*, North Carolina State University, Raleigh, NC. It has been 30 years since the discovery that persistent organic pollutants (POPs) such as PCBs and DDT are distributed globally. This global distribution is controlled by distillation, condensation, and selective retention/degradation as a function of latitude (temperature) and specific geophysical characteristics (ocean-air exchange versus soil-air exchange). We have investigated the latitudinal changes in concentrations and relative abundance of various POPs at different trophic levels in both remote and near-coastal areas of the Atlantic Ocean. We also have summarized data from the past 15 years for Massachusetts Bay. Our data support the hypothesis that POPs with high vapor pressure are migrating toward the poles. However, there appears to be a decoupling of the near-coastal pelagic system with the rest of the Atlantic, with near-coastal pelagic areas following a first order loss in PCB concentrations, and near-coastal benthic systems and the open ocean following a more gradual decline. The decoupling of trophic levels also is evident, with lower trophic levels reflecting current exposure in the water and higher trophic levels reflecting historical exposure. We hypothesize that reservoirs of POPs in sediments and in the atmosphere are buffering changes in benthic and open-ocean systems, respectively. The implications are that consumers of near-coastal and lower-trophic level organisms will experience a more rapid decline in xenobiotic exposure than consumers of benthic and open-ocean organisms. We have made new estimates of hemispherical transport rates and latitudinal residence times for several POPs. We use these estimates to predict future exposure and body burdens in humans and three different marine mammals: the white-sided dolphin, the pilot whale, and the northern right whale.

316 Modelling the Trophodynamics of Organic Contaminants in the Arctic Marine Food Web. Hickie, B.E.* Trent University, Peterborough, ON, Canada; B. Hargrave, Dept. Fisheries & Oceans, Dartmouth, NS, Canada; D.C.G. Muir, Environment Canada, Burlington, ON; D. Mackay, Trent University, Peterborough, ON, Canada. Despite the long distances from developed regions, many persistent organic pollutants (POPs) accumulate in Arctic ecosystems and biomagnify through the marine food web to marine mammals such as beluga whales (*Delphinapterus leucas*) and ringed seals (*Phoca hispida*). While the concentrations of most POPs in Arctic marine mammals are considerably lower than in other areas (e.g Baltic Sea, St. Lawrence Estuary), the levels of several POPs are sufficiently high to be a health concern for the northern people who rely on them as a food resource (eg. PCBs, chlordane, toxaphene, DDTs). Bioaccumulation of POPs through the Arctic marine food web has been characterized in a year-round study of the lower food web (water, particles, phytoplankton, ice algae, invertebrates, Arctic cod) and by monitoring programs for marine mammals. Results from these studies were modelled using a fugacity-based steady-state food web model for the lower trophic levels and species-specific dynamic models for ringed seal and beluga. The combined models adequately predicted the bioaccumulation of most POPs examined, but tended to underpredict the bioaccumulation of ΣDDT and Σchlordane. Low predicted values may be attributed to seasonal shifts in food web dynamics, contaminant bioavailability, and to difficulties in measuring the low concentrations of many POPs in seawater (e.g. ΣDDT ~ 2 pg/L). The models can be used in reverse to calculate target concentrations of POPs in sea water (or other more easily sampled environmental media) that will ensure that marine mammals and other marine food resources will not exceed tolerable limits for human consumption.

317 Persistent Organochlorines (OCs) in Marine Mammals in the North American Arctic: Spatial and Temporal Trends and Hepatic CYP450 Activity. Muir, D.*, Environment Canada, NWRI, Burlington, ON; Macdonald, C., Northern Environ. Services, Pinawa MB; Weis, M., University of Windsor, Windsor ON; Becker, P., NIST, Charleston SC; Schantz, M., NIST Gaithersburg MD USA; Lockhart L., and Metner, D. Fisheries & Oceans, Winnipeg MB. From 1990 to 1996 samples of two resident arctic species, the ringed seal (*Phoca hispida*) and beluga (*Delphinapterus leucas*) were collected, with the help of aboriginal hunters, in the Canadian arctic and Alaska to examine spatial and temporal trends of persistent OCs in these populations and, by inference, their food webs. Liver samples from Beaufort Sea beluga and W. Hudson Bay ringed seals was frozen on dry ice and later assayed for CYP450 mixed function oxidase (EROD) activity. Beluga from Cook Inlet were distinguished from all other groups by having the lowest levels of major OCs (toxaphene, chlordanes, total DDT, total PCB congeners (sPCB)). For e.g., mean sPCB levels in Cook Inlet males (2600 ng/g) were about half the levels in males from the S. Beaufort Sea population (5010 ng/g). Ringed seals from 7 locations in the east/central Canadian arctic (W. Hudson Bay to S. Ellesmere Is) had similar concentrations of major OCs after adjustment for effects of age, sex and especially blubber thickness. TEQs due to non-ortho-PCBs were significantly correlated with EROD activity in liver of male seals ($R^2 = 0.54$, $P<0.001$) but not in females ($P>0.1$). A potentially significant confounding factor is that sPCBs in male seals were significantly correlated with age (R^2=0.79). However, CB126, the major contributor to TEQs, was not strongly correlated with age (R^2=0.1; P=0.13). Therefore, the correlation of nPCB TEQs with EROD appears to be relatively unaffected by age although, for reasons yet to be explained, it is greatly influenced by the sex of the animals.

318 Selective Inhibition of *In vitro* Metabolism in Hepatic Microsomes to Determine the Specific Cytochrome P450 Isozyme Activities Towards the Biotransformation of PCBs, PCDDs, and PCDFs in Harbour Seal (*Phoca vitulina*). Letcher, R.J. *, Boon, J.P. and Lewis, W., Netherlands Institute for Sea Research (NIOZ), Texel, The Netherlands; van den Berg, M. and Seinen, W., Utrecht University (RITOX), Utrecht, The Netherlands. The role of cytochrome P450 (CYP) 1A-, CYP2B- and CYP3A-type isozymes in the metabolism of PCBs and PCDD/Fs in the hepatic microsomes of harbour seal was investigated using an *in vitro* PCB metabolism bioassay in the presence of compounds known to inhibit specific microsomal CYP isozymes in humans and rats. In the absence of inhibitors, PCB-15 and PCB-77 underwent significant metabolic depletion from a twelve PCB congener mixture. Ellipticine, a CYP1A-type inhibitor, showed a significant concentration-dependent inhibition of PCB-15 and -77 metabolism at ca. 0.25 :M, with ca. 80% maximal inhibition at ca. 1.0 :M and ca. 20% at ca. 0.5 :M, respectively. The CYP3A-type ketoconazole inhibited PCB-15 and -77 metabolism at a maximum of 90% and 40%, respectively, at ca. 10 μM. Both CYP1A- and CYP3A-type enzymes appeared to be responsible for coplanar PCB metabolism. In a separate mixture, the metabolic depletion of PCB-15 and -77, 2,7-Cl_2-CDD and 1,2,3,4,8-Cl_5-CDF could be completely inhibited with ellipticine (20 μM). For ketoconazole, inhibition of 80% and 85% of the metabolism occurred for PCB-15 and 2,7-Cl_2-CDD, respectively, and 44% and 48% of the metabolism for PCB-77 and 1,2,3,4,8-Cl_5-CDF, respectively. For CYP3A-type biotransformation mediation *in vitro,* dioxin/furan and coplanar PCB metabolism may be contingent on the degree of chlorination, and not on an obligatory low energy barrier to phenyl ring rotation. The profile of CYP isozyme activity responsible for coplanar PCB, PCDD, and PCDF metabolism *in vitro* was partially defined for harbour seal.

319 Environmental Contaminants and the Risk of Adverse Effects in Marine Mammals: an Overview. Ross, P.S., Institute of Ocean Sciences, Sidney, BC Canada. Despite the implementation of regulatory controls in the 1970s, marine mammals in many parts of the world continue to be exposed to elevated concentrations of polychlorinated biphenyls (PCBs), dioxins (PCDDs), furans (PCDFs), the DDT family, and numerous other pesticides. While conclusive evidence that contaminants are adversely affecting free-ranging marine mammals is still lacking, an accumulated "weight of evidence" suggests that ambient environmental (dietary) levels of contaminants have caused reproductive impairment, endocrine disruption and skeletal malformations in different areas. This epidemiological or observational evidence is supplemented by i) captive feeding studies, where harbour seals fed contaminated fish have suffered from reproductive impairment, immunotoxicity and endocrine disruption; ii) simplified laboratory rodent studies, which have identified chemicals of concern and the mechanism of their effects; and iii) parallel seal-rodent studies, where rats have been exposed to the same dietary contaminant mixture as the harbour seals in captive feeding studies (Atlantic or Baltic Sea herring). Taken together, such studies provide a means of assessing the risk of contaminant-related effects in free-ranging marine mammal populations. In this regard, the harbour seal, Phoca vitulina, has become a useful sentinel species for marine mammals. This pinniped is widely distributed throughout the northern hemisphere, was the victim of a high-profile mass mortality in Europe in 1988, and has been extensively studied. While direct evidence is likely to remain elusive as a result of the difficulties in carrying out toxicological studies in marine mammals, the potential risks presented by environmental contaminants may be estimated by this "weight of evidence", combined with knowledge of contaminant levels in a particular marine mammal population. In this regard, contaminants, and particularly PCBs, in harbour seals inhabiting much of northern Europe, Puget Sound, San Francisco Bay, and parts of British Columbia, appear to be high enough to present a risk of adverse effects. Studies being carried out in different parts of the world suggest that many persistent contaminants continue to cycle in the marine environment, and will continue to present a risk to marine mammals for many years to come.

320 Susceptibility to Infections and Immune Status in Inuit Infants Exposed to Organochlorines. Dewailly, É.*, Ayotte, P., Bruneau. S., Laliberté, C., Gingras, S., Quebec Public Health Center (CHUQ), Beauport, Québec, Belles-Isles, M., Roy, R., CHUQ Research Center, Ste-Foy, Québec, Canada. The influence of pre- and postnatal exposure to organochlorines on the immune status and the incidence of infections was investigated in Inuit infants from Nunavik (Arctic Québec). A total of 171 mother-infant pairs were seen at least once during the 12-month follow up period (98 breast fed and 73 bottle fed). Concentrations of selected organochlorines in breast milk fat were used as prenatal exposure surrogates. Postnatal exposure was approximated by multiplying organochlorine levels in milk fat by the breast feeding duration. Episodes of infections were compiled during the first year of life. Granulocytes, monocytes, lymphocytes and immunoglobulins were measured in venous blood at 3, 7 and 12 months of age. From 0 to 3 months of age, the risk of otitis media among breast fed infants appeared lower than in bottle-fed infants (RR = 0.70, 95%-CI = 0.47-1.03), but not later during the first year of life. During the second (4-8 months) and third (9-12 months) follow-up periods, the risk of contracting otitis media in infants receiving the highest doses of organochlorines through breast feeding was greater than that of infants receiving the lowest doses (ex.: for hexachlorobenzene at 9-12 months: RR = 2.24, 95%-CI = 1.21 - 4.16). Cumulative PCB exposure through breast feeding (r = -0.27; p 0.05) and breast feeding duration (r = -0.31; p 0.05) were inversely correlated to the CD4/CD8 ratio during the second follow up period. We conclude that organochlorine exposure through breast feeding can modulate the immune system function and may be a risk factor for acute otitis media in Inuit infants.

321 Extraction and Isolation of Linear Alcohol Ethoxylate from Fish. Tolls, J.*; Haller, M., RITOX, Utrecht, NL; Sijm, D.T.H.M., RIVM, Bilthoven, NL. The development of an analytical method for determination of alcohol ethoxylate (AE), the most important nonionic surfactant in household detergents, in fathead minnows is described. Combination of matrix solid phase dispersion (MSPD) extraction from fish with existing cleanup methods renders possible the determination of the AE concentrations in bioconcentration experiments. The method development as well as the bioconcentration experiments were performed with simple mixtures of individual constituents of AEs. Recoveries of the analytes were higher than 75% for water as well as for fish samples. AEs incurred during bioconcentration could be quantified without integration being compromised by interferences for all compounds except for $C_{14}EO_{14}$. The limit of quantitation for individual AE constituents was estimated to be 1.5 nM and 150 nmole x kg^{-1} in water and fish, respectively. Parallel determinations of the concentrations of ^{14}C-$C_{13}EO_8$ in fish by TLC-RAD and liquid scintillation counting and by HPLC measurements after isolation and derivatization yielded very similar results demonstrating the validity of the isolation and measurement method.

322 Quantitative Determination of Alcohol Ethoxylate Surfactants in Marine Sediments by Electrospray LC/MS. D. Shang, M. Ikonomou and R.W. Macdonald, Institute of Ocean Sciences, Sidney, B.C. V8L 4B2 Canada. Over the past few years our interest has been the development of analytical methodology for the determination of non-ionic surfactants in environmental samples. We have developed a highly specific analytical method, based on normal phase LC/ESI-MS, for the analysis of nonylphenol polyethoxylates in marine sediments (see SETAC 97. PO993). In this work we have extended our method to another group of non-ionic compounds, *viz.* alcohol polyethoxylates (AEOs). This procedure involves extraction of marine sediment using Accelerated Solvent Extractor (ASE) followed by clean-up on cyanopropyl (CN) silica solid phase extraction (SPE) cartridges. Optimal chromatography separation was achieved by using normal phase LC to elute all the oligomers of AEO. The LC was coupled to the MS via a mega-flow-ESI probe which required no splitting of the LC effluent. The high volatility solvents used in normal phase LC permitted for the entire LC effluent, 0.2 ml/min, to be handled by the mega-flow-ESI interface. For this particular application this arrangement provided superior reproducibility and sensitivity in comparison to the results obtained from the conventional ESI probe where only 10 to 20μl/min were delivered to the ESI tip. Addition of sodium acetate to both sample solvent and mobile phase facilitated stable ESI operation and detection of all the AEOs as sodium adducts. The high specificity of the LC-ESI/MS technique enables interference free quantitation of AEO species at part-per-billion concentration levels. The method can distinguish highly branched AEOs from isomeric linear ethylene-based AEOs. Spike recovery was used to validate the analytical method. This method was used to determine the concentrations of AEOs in the sediments from Strait of Georgia, B.C.

323 Phthalate Esters in Environmental Samples: A Review on Analytical Methodology and Toxicity. Lin, Z. P.* [1,2], Michael G. Ikonomou[2] and Frank Gobas[1], [1]School of Resource and Environmental Management, Faculty of Applied Sciences, Simon Fraser University, Burnaby, BC, Canada; [2]Ocean Chemistry, Institute for Ocean Sciences, Sidney, BC, Canada. Phthalate esters are produced in very high volumes (2,700,000 tones/year) and have widespread use in many industrial applications particularly in the plastics industry. Because of the high volumes produced the environmental fate and toxic effects of phthalate esters have been of concern to environmental chemists (ECPI 1996) and toxicologists (Staples 1997) for many years. Based on reports published in the open literature it seems that the scientific interest on the subject has been going though cycles since the early 70s. Recently the environmental impact of phthalates has been of high importance to the environmental scientific community and a whole session was devoted to the subject during SETAC-96. We have been working towards the development of a statistical model that could be used to predict the distribution of phthalates in environmental matrices such as water, sediments and biota provided that the input sources are characterized. This prompted a review of the literature especially on the analytical methods used for the determination of phthalates since a substantial number of samples will have to be analyzed to provide data for validating the model. In this presentation we will present a summary of the advances made for the determination of 14 of the most common phthalate esters in environmental samples over the last 25 years. Data on toxicity and mutagenicity of these phthalates will also be reviewed.

324 Fate of Toluenediisocyanate and Methylenediphenyl Diisocyanate in the Aquatic Environment. Bailey, R. E.*, Bailey Associates, Midland, MI; Yakabe, Y., Chemicals Inspection and Testing Institute, Kurume, Japan; Ode, R. H., Bayer Corporation, New Martinsville, WV; Pemberton, D. and Tury, B., Gilbert International, Ltd., Manchester, U. K. The reactions of toluenediisocyanate (TDI) and polymeric methylenediphenyl diisocyanate (pMDI) with water, under conditions relevant to environmental contact, have been studied. These diisocyanates are major intermediates in the production of polyurethanes, with global production of each greater than 1 billion kg per year. Previous literature has focused primarily on isocyanate reactivity during polyurethane production rather than in the aquatic environment. Both TDI and pMDI are hydrophobic and react heterogeneously with water, the observed rate being dependent on the rate of stirring. Although with rapid stirring a half-life of a few minutes is observed for TDI, and of the order of 20 hours for the considerably more viscous pMDI, unstirred reactions are considerably slower and half-lives of six days or longer are found for both materials. The products of reaction with water are predominantly solid polyureas, at all but very low loadings of diisocyanate, with low concentrations of diamine (toluenediamine or methylenedianiline) in the aqueous phase. The polyurea products are highly insoluble and appear to be inert under environmental conditions. Production of diamine is minimized with poor agitation of the reaction mixture. These studies are consistent with the minimal effects noted from the accidental spillage of TDI and pMDI in the environment.

325 Use of Source-Receptor PAH Diagnostics to Explain the Spatial and Temporal Variations Observed in Massachusetts Bay Sediments. Graham, S.E.*, Shea, D., North Carolina State University, Raleigh, NC. Trace level concentrations of over 40 polycyclic aromatic hydrocarbons (PAHs) were determined in sediment cores and sediment traps collected in Massachusetts and Cape Cod Bays. PAH diagnostic techniques, including parent compound isomer ratios (PCIR) and parent compound distribution (PCD) analysis, were employed to help identify PAH sources and important fate processes. Significant variation in the fractional abundance of anthracene (ϕ_a) in the buried sediment has been successfully modeled (R^2=0.67, $p<0.0001$) using multiple linear regression (MLR) with site location, isomer concentration, and organic carbon comprising the explanatory variables. These data suggest changes in the anthracene (ANT) to phenanthrene (PHE) ratio were related to both pre- and post-depositional processes. Decreases in ϕ_a during atmospheric particulate transport probably occur because of the increased photolytic susceptibility of ANT relative to PHE, as evidenced by low ϕ_a at distant sites. Furthermore, we observed a decrease in ϕ_a with depth in all sediment cores. This probably reflects the preferential desorption of PHE relative to ANT caused by differences in their apparent water and organic-carbon solubilities. Similar results were obtained for other PCIRs such as the benzo[a]pyrene-to-benzo[e]pyrene ratio. A decrease in the input of combustion-related PAH in recent years could play a minor role in the decrease of all combustion-related fractional abundances, although the fractional abundance of the combustion-indicative benzofluoranthanes-to-benzo[e]pyrene PCIR (with similar solubilities) shows no relationship with the same explanatory variables.

326 Analysis of Urban Particulate Standard Reference Materials for the Determination of Chlorinated Organic Contaminants and Additional Chemical and Physical Properties. Poster, D.L.*, Schantz, M.M., Wise, S.A., Vangel, M.G. National Institute of Standards and Technology, Gaithersburg, MD. A previously issued National Institute of Standards and Technology (NIST) Standard Reference Material, SRM 1649, Organics/Urban Dust, which is composed of urban air particulate matter that was collected in the Washington D.C. area in the late 1970's, has been analyzed for chlorinated organic contaminants (polychlorinated biphenyls (PCBs) and chlorinated pesticides) to provide certified values for a new class of compounds relative to the former certification. The material will be reissued as SRM 1649a. Four different analytical techniques were used for this work. Specifically, two different methods of extraction (Soxhlet and pressurized fluid extraction) were used in conjunction with sample analysis by gas chromatography with two different columns (5 % phenyl-methyl polysiloxane and 50 % methyl C-18 dimethyl polysiloxane) that exhibit distinct selectivity, and with two different modes of detection (electron capture detection and mass spectrometry). The results from these techniques were combined to generate certified concentrations for 29 PCB congeners and 8 chlorinated pesticides. Ancillary assessments of additional chemical and physical properties of SRM 1649a include homogeneity, moisture, total organic carbon, extractable mass, and the particle-size distribution. The approach and the results for the certification of the PCB congeners and chlorinated pesticides in SRM 1649a, and the determination of the additional chemical and physical properties will be presented. In addition, the analysis of SRM 1648, Urban Particulate Matter, for the determination of PCBs and chlorinated pesticides will also discussed.

327 Characterization of Enantiomeric Ratios of Chiral Agrochemicals in Environmental Standard Reference Materials. Poster, D.L.*, Lagarde, F., Wise, S.A., National Institute of Standards and Technology, Gaithersburg, MD. Measurements of enantiomeric ratios of agrochemicals that contain chiral centers can be used as environmental tracers and as possible indicators of chemical toxicity. We have analyzed several solution, biological, and marine sediment Standard Reference Materials (SRMs) for a range of optically active agrochemicals, including α-hexachlorocyclohexane, *cis*- and *trans*-chlordane, heptachlor, and the oxygenated metabolites heptachlorepoxide and oxychlordane, in an effort to provide values for many of these compounds in solution and natural matrix environmental SRMs. Measurements were accomplished through the use of chiral gas chromatography equipped with electron capture detection or mass selective detection. Chiral-phase capillary GC columns, including permethylated α-, β-, and γ-cyclodextrin, were used to separate enantiomers. A *tert*-butyl dimethyl silyl β-cyclodextrin was also used, as well as a combination of non-chiral and chiral phase capillary GC columns. Racemic and enriched standards were used to determine enantiomeric ratios. The results of these analyses will be presented, along with an emphasis on the methods of analysis as an approach for the determination of enantiomer values in environmental SRMs.

328 Speciation Measurement of Cobalt and Chromium Interacting with (Amino) Carboxylic Acids. McArdell, C.S.*,1, Stone, A.T.2, Buerge, I.J.1 and Hug, S.J.1, 1Swiss Federal Institute for Environmental Science and Technology (EAWAG) & Swiss Federal Institute of Technology (ETH), Duebendorf, Switzerland and 2Department of Geography and Environmental Engineering, The Johns Hopkins University, Baltimore, MD, USA. Information on the chemical form (speciation) of an element is of fundamental importance to assess its environmental impact. Capillary Electrophoresis (CE) can be used to separate and directly determine metal complexes. Reactions of EDTA, NTA, and related aminocarboxylic chelating agents with CoIIIOOH(s), representing the sorbed contaminant metal CoIII, are performed to study source and sink terms for mobile CoIII complexes in subsurface environments. CoOOH reacting with EDTA and NTA yields products reflecting both ligand-assisted dissolution (i.e. soluble CoIII species) and reductive dissolution (CoII and aminocarboxylate oxidation products). Aminocarboxylate oxidation occurs primarily through N-dealkylation. Another study deals with chromium speciation. Reduction of the toxic CrVI to essentially non-toxic CrIII in the environment often occurs through reaction with FeII. This reaction is strongly influenced by pH and organic ligands. In reactions of CrVI with FeII in presence of oxalate, formation of CrIII complexes with oxalate were observed by analysis with CE. Stability constants of CrIII with organic ligands such as oxalate, salicylate or citrate are not known. CE measurements enable differentiation and semi-quantification of these complexes in different ratios between metal and ligand. Reaction rates are estimated in kinetic experiments for the formation of CrIII(oxalate)1 and CrIII(oxalate)2.

329 Occurrence of the Fluoroquinolone Antibiotic Ciprofloxacin in Hospital and Municipal Wastewaters. Alder, A.C.[1]*, Golet, E.[1], Hartmann, A.[2], Ibric, S.[1], Suter, M.J.-F.[1]; [1]Swiss Federal Institute for Environmental Science and Technology (EAWAG), CH-8600-Dübendorf, Switzerland and [2]Swiss Federal Institute of Technology (ETH), CH-8092 Zurich, Switzerland. There is increasing evidence for the presence of substantial amounts of pharmaceutical drugs in the aquatic environment. At present, the ecological consequences of these low-level drug exposures are unclear. Fluoroquinolone (FQ) antibiotics, inhibitors of the bacterial DNA unwinding enzyme gyrase, are important broad spectrum antibiotics licensed for the use in both humans and animals. The concentration of the leading FQ in human medicinal use, Ciprofloxacin, was determined by reversed-phase HPLC with fluorescence detection in hospital wastewaters and qualitatively confirmed by electrospray mass spectrometry. Concentrations ranged from 5 to 100 μg/L. Ciprofloxacin was found to be bioeliminated to 56± 11% in 24 days using a modified OECD 302B in hospital wastewater. Bioavailability was unexpectedly high, as indicated by a bacterial genotoxicity assay (umuC test), which is highly sensitive for FQ antibiotics. Therefore, we expected substantial amounts of bioavailable FQ antibiotics in wastewaters. An enrichment procedure had to be developed for municipal wastewaters. Acidified aqueous samples were extracted with cationic exchanger disks, which were subsequently eluted with basic methanol. The concentrations of Ciprofloxacin in 24 h composite samples in municipal wastewaters ranged from 0.2Ð0.4 μg/L in primary effluents and 0.1 μg/L in secondary effluents. The elimination of Ciprofloxacin from the wastewater stream during aerobic treatment varied from 55Ð75%. A general LC-MS method for the confirmation of our results is under development. Our findings indicate that fluoroquinolones are present in wastewaters in concentrations which, in combination with other antibiotics, might have effects on some of the most susceptible microorganisms and could contribute to resistances in human pathogenic microorganisms.

330 Removal of Fragrance Materials During Sewage Treatment. Simonich, S.L.*, Begley, W.M., Eckhoff, W.S., Bernhard, M.J., The Procter & Gamble Company, Cincinnati OH, USA. Fragrance materials (FMs) are used in a wide array of consumer products, at low concentrations. Analytical methods were developed to measure representative FMs, with a wide range of physical-chemical properties, in influent, primary effluent, and final effluent of sewage treatment plants. Analytical methods include the use of large volume solid phase extraction, accelerated solvent extraction of solids, and perdeuterated FMs as internal standards. The physical-chemical properties (octanol-water partition coefficient and Henrys Law Constant) of the group of FMs ranged over several orders of magnitude. Fragrance material removal during activated sludge and trickling filter sewage treatment was measured. Measured removal after primary settling ranged from 32 - 67%, while measured final removals ranged from 80 to greater than 99%, depending on the physical-chemical properties and biodegradabilities of the FM. Measured final removals from activated sludge sewage treatment plants were slightly higher than measured final removals from trickling filter plants. Various sewage treatment plant models (SimpleTreat, TOXCHEM+, and ASTREAT) were used to estimate removal of fragrance materials, based on their physical-chemical properties, and compared to measured removals. In general, these models appear to underestimate fragrance material removal during sewage treatment.

331 Applying Ecosystem Valuation to Regional Ecological Risk Assessment and Management. McLaughlin, J.F.*, Landis, W.G., Institute of Environmental Toxicology and Chemistry, Robbins, L.A., Huxley College, Western Washington University, Bellingham, WA. We developed a method to determine how anthropogenic impacts should be managed across a region. The method uses socially defined ecosystem values to define objectives for regional risk assessment. The method consists of four phases: (1) identifying desired ecosystem responses, (2) quantifying stressor-response relationships at the regional level, (3) determining necessary constraints on anthropogenic stressors, and (4) implementing regional risk management. The first phase identifies ecosystem responses of greatest concern using four social survey instruments: literature searches, key informant interviews, focus groups, and public surveys. The second phase develops mathematical models of stressor-response relationships to predict when responses identified in phase one will exceed socially tolerable limits. The models are developed using methods of time-series analysis applied to data on relevant economic, ecological, and climatic variables. The third phase applies these models to determine constraints on human impacts necessary to prevent ecosystem responses from exceeding tolerable limits. The final phase presents results of the analysis to public, commercial, and agency stakeholders on an individual basis and through public forums. Subsequently, stakeholders participate in developing strategies to implement regional risk management. Our approach integrates social and environmental sciences to conduct a regional risk assessment. It incorporates three factors necessary for successful risk management: (1) relevance to stakeholder values, (2) a quantitative understanding of stressor-response relationships, and (3) stakeholder participation. We will illustrate our method and its application to several regional ecosystems in the Pacific Northwest.

332 Integrated Decision Analysis Tools for Watershed Management. Breckenridge, R. P.*, Lockheed Martin Idaho Technologies, Idaho National Engineering and Environmental Laboratory, Idaho Falls, ID, Benjamin, L., Henry's Fork Foundation, Ashton, ID, Lall, U, Utah State University, Logan, UT, and Minshall, G. W., Idaho State University, Pocatello, ID. Integrated decision analysis tools that can be used to answer land management and planning questions for entire watersheds and ensure that local, regional, and national issues are addressed are being developed by a collaborative team of the INEEL, Utah State University, Idaho State University and the Henry's Fork Foundation. The approach utilizes data from State and Federal agencies, academic institutions, and others with selected models and displays modeling results using a geographic information system (GIS). Major components include data management, hydrologic modeling of surface and groundwater, land use and habitat assessment, socio-economic assessment, and education (local schools assistance in data collection, and communication and education of the public on watershed issues). Output from this system is being made available to users and decision makers via the Internet using MapObjectsÒ. Users will be able to query data sets, produce maps of resource information in the watershed, evaluate options, and assess how changes in population, land use, and other human activities will impact resources in their watershed. Decisionmakers, including local, state, and federal agency personnel, private land owners, and developers are closely involved in the development of requirements and questions needed to support management of ecosystem processes at the watershed level. This process is initially being developed for the Teton watershed in Idaho and Wyoming. The universal framework being used will be transferable to other watersheds and will enhance the ability of Federal and State agencies and Universities to partner with local communities to develop and implement land management strategies at the watershed level.

333 A Data Screening Tool For Watershed Ecological Risk Assessment To Evaluate Complex Multi-Disciplinary Monitoring Programs. J.E. Babendreier*, D.L. Gallagher, Department of Environmental Engineering, D.S. Cherry, R.J. Currie, H.A. Latimer; Department of Biology, Virginia Tech, Blacksburg, VA. Tasks associated with data management of complex environmental systems are often underestimated and can remain a limiting factor during analysis. Ecological risk assessment incorporates multi-disciplinary monitoring approaches, and quantitative relationships between various endpoints investigated are not usually well defined or even suspected. This work describes the development and application of data management tools for ecological risk assessment of the Leading Creek Watershed in southeastern Ohio. Task areas of site monitoring, data management and endpoint integration were broken down and normalized into 13 data endpoint groups for comparison: Biodiversity, Biomass, Sediment Toxicity, Sediment Chemistry, Acute Toxicity, Water Column Chemistry, (In-situ) Clam Toxicity, RBP Habitat, Riparian Zone, Sediment Depth (bed), Land Use/Cover, Channel Hydraulics, and Flow. Data organization was accomplished through an integrated database linked to a GIS. The database field definition and structure was first normalized for all data sets defined. System design also allowed for calculation of additional system variables representing statistics of other variables within a specific group (spatial or temporal averaging, etc.). Query design was then used to summarize the data into paired sets of multiple columns representing a final list of 128 system variables initially selected for screening. Finally, macro programming was undertaken to allow for single-key access/query capability to automatically rank matched data sets and generate graphics. Pearson correlation coefficients were used as an initial screening tool for ranking variable sets. As an example, for screening of the Leading Creek project data, this analytical tool allowed for the rapid identification of the 325 most significant relationships (R^2>0.25) out of the 8192 possible unique relationships among 128 variables studied.

334 TMDLs for Narrative Standards: Convergence with Ecological Risk Assessment. Butcher, J.B.*, Tetra Tech, Inc., Research Triangle Park, NC; Creager, C.M., Tetra Tech, Inc., Calistoga, CA; Craig, J.P., Tetra Tech, Inc., Fairfax, VA. Total Maximum Daily Loads (TMDLs) are required by the Clean Water Act for water quality-limited waters where technology-based controls are insufficient to achieve standards. The TMDL consists of an allocation of loading capacity to point and nonpoint sources. The TMDL is straightforward for pollutants with numeric water quality standards, but is more difficult for stressors for which there are not numeric standards, but which cause impairment of narrative water quality standards, such as general narrative specifications for the support of aquatic life. We have proposed methods to address narrative-standard TMDLs in a series of draft protocols for EPA. These protocols draw upon ecological risk assessment theory, but take a broader perspective in addressing both ecological and non-ecological designated uses in a regulatory context. For instance, excess loads of sediment may result in degraded fish habitat, decreased recreational opportunities, degraded drinking water supplies all potential impairments of designated uses. Many such waters are clearly impaired, but developing TMDLs presents problems because both the waterbody loading capacity for sediment, which is highly site-specific, and sediment loading rates, which are highly variable in time, are difficult to determine quantitatively. To address the difficulties narrative-standard TMDLs, the protocols advocate development of numeric indicators of instream impact and methods to link indicators to land use practices in the watershed. These indicators are analogous to measures of effect associated with assessment endpoints in ecological risk assessment, but are more generally defined to encompass ecological measures, stressor measures, and impacts on non-ecological designated uses. This new paradigm can successfully be used to complete TMDLs, as exemplified by several recently-approved TMDLs for sediment, temperature, and nutrients.

335 Phosphorus and Reservoirs: a TMDL Is Not One Number. Canton, S.P.*, Chadwick Ecological Consultants, Littleton, CO; C. Paulson, Brown & Caldwell, Denver, CO. TMDLs are required under EPA regulations to protect a water body's designated uses. While a TMDL appears straightforward, it can be problematic for reservoirs. In 1985, standards were set for Cherry Creek Reservoir, CO: seasonal mean of 15 μg/L chlorophyll to be protected by a standard of 35 μg/L total phosphorus. This inlake TP standard was expanded to a TMDL of 14,270 lbs/yr. Monitoring data have shown the lake to be meeting the chlorophyll goal with an inlake phosphorus of roughly 60 μg/L and loads ranging from 5,000 lbs/yr to over 10,000 lbs/yr. Attempts to validate the 1985 TMDL loading estimate using 1992-97 data were unsuccessful. Review of TP loading models determined that no model could predict any individual year with certainty (but could predict the *average* condition). This appears to be a result of variation in internal loading events related to biological processes and climatic variation. This analysis pointed out a problem with the TMDL paradigm. While an allowable load is possible to compute for the Reservoir, it cannot based on a maximum amount, or even a daily load. Rather, it is an average allowable annual load, that includes a range of loads. The resulting recommended changes in inlake standards account for this annual variability, with standards based on 5-year rolling averages (including exceedence criteria and corrective actions). Rather than attempting to calculate a TMDL, per se, phosphorus loading recommendations were based on protecting these average inlake conditions under predicted future conditions in the watershed, with the understanding that the load will vary around an average in response to climatic conditions.

336 Total Maximum Daily Load [TMDL] Development and Agricultural Nonpoint Source Pollution. Bramblett, Jimmy R. USDA-NRCS, Athens, Georgia. The 25th anniversary of the Clean Water Act in October 1997 signaled a fundamental shift from point source contributors to nonpoint source contributors with respect to water quality impairments. Agriculture is often cited as the major polluter in the nonpoint source area. However, recent events in Georgia are bringing into question the validity of this paradigm. Litigation via the Sierrra Club has lead to a federal court order which is hastening TMDL development in Georgia. The Natural Resources Conservation Service is documenting US Environmental Protection Agency's [EPA] overestimates of agricultural nonpoint source pollution in TMDL development. The magnitude of EPA overestimates clearly demonstrates the need for agricultural expertise in TMDL development. A methodology has been developed by NRCS in Georgia to provide appropriate input relative to the nutrients, bacteria, chemicals, metals, and other constituents contained in runoff associated with agricultural operations. EPA has embraced this methodology for future TMDL development in Georgia.

337 Molecular Tracers of Organic Matter Sources to Surface Waters. Standley, L. J.*, Kaplan, L. A., Stroud Water Research Center, Academy of Natural Sciences, Avondale, PA. Sources of organic matter to ground and surface waters are of concern to the drinking water industry due to the necessity of treating source waters to remove natural and anthropogenic organic matter (OM) to prevent regrowth of pathogenic microorganisms. However, treatment can produce disinfection by-products that are toxic. Thus, the drinking water industry is interested in promoting best management practices in their source watersheds as a way of reducing their use of disinfectants. We have investigated the applicability of various compounds for use as molecular tracers of OM sources in surfaces waters from twenty watersheds in the United States, Canada, and Spain. Anthropogenic sources of OM include waste water treatment plants (WWTP), and feed-lot and urban runoff. Molecular tracers of these sources include fecal sterols, perfumes, caffeine, surfactant breakdown products (nonyl phenol), polycyclic aromatic hydrocarbons (PAH), *n*-alkanes, unresolved complex mixtures (UCM, petroleum and combustion derived), and veterinary medications. Four liter samples were filtered through precombusted glass fiber filters and then extracted using C-18 Empore disks. Molecular tracers and surrogate standards were quantified using gas chromatography/mass spectrometry with selected ion monitoring to improve sensitivity. Tracers associated with WWTP effluent, including fecal sterols, perfumes, caffeine, and surfactants, co-occurred in dissolved and particulate fractions of water samples, perfumes, caffeine, and nonyl phenols occurred primarily in the dissolved fraction, and fecal sterols were associated primarily with particulates. Urban tracers such as UCM and PAH also co-occurred in samples, with more water soluble PAH and UCM present primarily in the dissolved fraction and larger, less soluble PAH present in particulates. These results will be correlated with land-use patterns in target watersheds.

338 Quantification of Spatiotemporal Variability and Source Identification of Atmospheric Nitrogen Deposition in the Neuse River Basin (North Carolina). Whitall, D.R.*, Peierls, B.L., H.W. Paerl. University of North Carolina at Chapel Hill, Chapel Hill, NC. Showers, W.J. North Carolina State University, Raleigh, NC. Atmospheric deposition of nitrogen (AD-N), through both wet and dry deposition of NO_y, $NH_{3/4}$, and organics, may contribute >30% of the total *new* or externally supplied N to the Neuse River watershed. Excessive N loading to N-limited waters such as the Neuse has been shown to promote changes in microbial and algal community composition and function, harmful algal blooms, hypoxia/anoxia, and fish kills. In an ongoing study, we quantified the weekly wet and dry deposition of inorganic N at four sites (Goldsboro, Kinston, New Bern, Morehead City) on a northwest-southeast transect in the Neuse River basin since the summer of 1996. Current budget calculations indicate large spatiotemporal variability in N deposition, with the mean flux of N to the watershed via atmospheric sources estimated at 259 mg-N/m^2/year via NH_4 wet deposition, 58 mg-N/m^2/year via NH_3 dry deposition, 570 mg-N/m^2/year via NO_y wet deposition and 199 mg-N/m^2/year via NO_y dry deposition. Based on weekly sampling starting in June 1996, an estimated 13,000 metric tons of N/yr were delivered to the watershed, based on data from our 4 sites and the National Atmospheric Deposition Program site (NC41) in Wake County. Conservative estimates of watershed retention reveal that this makes up between 18 and 35% of the total N reaching the waterways of the basin. This information is essential for constructing a watershed level N budget. Preliminary stable nitrogen isotope measurements indicate the $\delta^{15}NH_4^+$ for New Bern and Kinston is between -2 and -4 per mil; this isotopic signal suggests that mobile sources rather than agricultural emissions are the dominant source of the NH_4^+ in the AD-N in the watershed.

339 Integrated Pest Management and Estuarine Aquaculture: New Opportunities and Challenges for Watershed Management. DeWitt, T.H.*, U.S. EPA, Newport, OR; Wellman, K.F., Battelle, Seattle, WA; Wildman, T., Independent Pest Management Consulting, Prosser, WA; Armstrong, D.A., University of Washington, Seattle, WA. Endemic thalassinid shrimp (*Neotrypea californiensis, Upogebia pugettensis*) are considered to be pests to the commercial production of oysters (*Crassostrea gigas*) because their burrowing buries and kills oysters. Oyster growers in two Washington state outer-coast estuaries have used the pesticide, carbaryl, since the 1960's to kill shrimp on their owned or leased tideflats. Presently, 800 acres of tideflats annually are sprayed with carbaryl in these estuaries. Oyster growers and regulatory agencies recently decided that integrated pest management (IPM) might be an appropriate approach for controlling the shrimp. IPM integrates population biology of the pest and crop species, susceptibility of pest life stages to various control methods, and knowledge of the pest density:crop damage relationship. It offers the potential to reduce the amounts of pesticide used and thus reduce non-target impacts. While widely practiced in terrestrial agriculture, IPM has not been used previously to control pests in an estuarine aquaculture environment. Our team evaluated whether existing information could be used to develop an IPM plan for burrowing shrimp. We found that critical knowledge is lacking (including shrimp population monitoring methods, shrimp population growth models, and non-chemical control methods) which precludes the immediate creation of an IPM plan. However, we believe that the concept has merit and that an IPM plan could be developed once the critical knowledge is obtained. As interest in estuarine and marine aquaculture increases, we expect that pest damage and attendant need for control strategies, such as IPM, will also increase. This case study illustrates unique challenges facing the application of IPM to aquaculture in estuarine environments.

340 Risk Assessment of Physical Stressors in CSO Discharges to an Urbanized Estuary. Wisdom, C.* Parametrix, Inc., Kirkland, WA, Volosin, J., Parametrix, Inc., Kirkland, WA, DeForest, D., Parametrix, Inc., Kirkland, WA, Cardwell, R., Parametrix, Inc., Kirkland, WA, Toll, J, Parametrix, Inc., Kirkland, WA, Munger, S., King County Department of Natural Resources, Seattle, WA, Strand, J., King County Department of Natural Resources, Seattle, WA, Simmonds, J., King County Department of Natural Resources, Seattle, WA. Discharges from combined sewer overflows (CSOs) can impact the environment through chemical, microbiological or physical effects. Of these, evaluating the risks to aquatic life from physical stressors presents a unique set of issues. This is because physical stressors have rarely been evaluated in risk assessments, which have traditionally focussed solely on chemicals released to the environment by human activities. In a study of the effects of CSO discharges on an urbanized estuary, several potential physical effects were identified. These effects are: increases in total suspended solids (TSS) and sedimentation; loss of benthic sediment habitat from scouring, reductions in water column salinity, dissolved oxygen; and pH, changes in water column temperature; and increases in water velocity displacing water column organisms. To identify risks to aquatic life from physical stressors, both effects thresholds and exposure parameters were defined as well as methods for characterizing risks for each stressor. Effects thresholds were developed from either existing regulatory criteria or standards or relevant data from the scientific literature. Exposure parameters were developed from either a field sampling program or from a hydrodynamic model of the estuary. Once effects threshold and exposure levels were defined, they were combined to characterize risks to aquatic life by either (1) calculating hazard quotients, or (2) probabilistically integrating them to evaluate the community level response. Additionally, physical effects can be manifested in the area directly around the CSO discharge as well as in areas farfield from individual discharges. Therefore, the spatial and temporal resolutions were identified for each stressor. Preliminary analyses of risks indicate no significant risk to the aquatic life community from TSS, sedimentation, DO, pH, or temperature related to CSO discharges. Nearfield effects of erosion and displacement were detected, as were decreases in salinity in farfield areas.

341 Avian Test Species Selection. P.J.Edwards and E.W.Schafer. The Avian Toxicity Workshop in Pensacola recommended the use of an Up and Down procedure to evaluate the approximate LD50 (ALD50). This test procedure will normally allow an ALD50 to be determined with very few animals (<10) and is appropriate for testing additional species to estimate the HD5 and remove the uncertainty factor in risk assessment applied for differences in species sensitivity. To determine the HD5 directly it is necessary to test at least 4-5 phylogenically unrelated species. The purpose of this paper is to recommend a short list of species from different Orders which are most appropriate for testing. This is not considered to be an exclusive list, but there are benefits in testing from a short list of species, although this is not necessary in principle for the determination of the HD5. Consideration for selection is given to 1) Phylogeny and relevance in agriculture; 2) Captive breeding and tolerance to laboratory conditions; 3) Availability; 4) Regurgitation; 5) Sensitivity; and 6) Size.

342 Development of the Canadian Tissue Residue Guideline for Methylmercury to Protect Wildlife Consumers of Fish. Miskimmin, B.M.*, Limnos Freshwater Consultants, Vernon, BC, Canada; Caux, P.-Y., and Kent, R.A., Environment Canada, Hull, QC, Canada. Methylmercury has long been recognized as the most toxic form of mercury in the environment. As part of an effort to develop nationally consistent, scientifically defensible tissue residue guidelines (TRG) to protect piscivorous birds and wildlife, methylmercury was considered to be a priority contaminant by the Canadian Council of Ministers of the Environment. Avian and mammalian wildlife that are dependent on aquatic biota such as fish or shellfish are susceptible to accumulating toxic levels of contaminants that biomagnify. In association with extensive fish tissue monitoring for mercury, effects-based criteria are required to help determine relative risk to vulnerable receptors. We comprehensively examined the available literature on the uses, chemical properties, sources, environmental behaviour, bioaccumulation and other aspects of methylmercury in the environment. We evaluated diet-based acute and chronic toxicity data for piscivorous avian receptors such as loons, herons and eagles, and for mammals such as mink and otter. The recommended TRG is based on tolerable daily intakes for avian and mammalian wildlife. The derivation of the final methylmercury guideline value is in progress, and will be presented.

343 Bioaccumulation and Effects of Metals from Mine Tailings in Small Mammals and Frogs. Champoux, L.*, Canadian Wildlife Service, Sainte-Foy, Québec, Trudeau,S., Canadian Wildlife Service, Hull, Québec, Gagnon, C., Soprin-ADS, Québec, Québec, Scheuhammer, A.M., Canadian Wildlife Service, Hull, Québec. Rouyn-Noranda, a major copper and gold ore region in northwestern Québec, contains a large number of sites contaminated with mining wastes. The present study was initiated to investigate the potential effects of environmental metal contamination from mine wastes on aquatic and terrestrial wildlife. From 1994 to 1996, small mammals (mice, voles and shrews) and frogs (green, mink and wood frogs) were collected from different sites in the area, and their tissues analysed for As, Cd, Cu, Ni, Pb, and Zn. Selected specimens were also analysed for biomarkers: δ-amino-levulinic acid dehydratase (ALA-d), porphyrins, and metallothionein. Histopathology was performed on some samples, and any physical deformities were also noted. Compared with values reported in the scientific literature, only a few individuals from the present study had high levels of metals. Voles and shrews from one of the sites had high blood ALA-d ratios, indicative of recent elevated Pb exposure. Metallothionein concentrations were low in all of the samples analysed. No neoplastic lesions were observed, but a few samples of vole hepatocytes had heterogenous size of cells and nucleus. Only one Mink frog out of 92 had a non-traumatic limb deformity. Although a final interpretation of the results awaits completion of analyses, results to date indicate that contamination and effects of metals from mine tailings in small mammals and frogs in the Rouyn-Noranda region are minor. However, based on inventories and trapping efforts, abundance and diversity of small mammals and amphibians appear to be reduced at the contaminated sites.

344 Persistent Organochlorine Residues in Oceanic Birds from Pacific and Indian Oceans. Guruge, K. S.*, Tanabe, S., Hashimoto, J., Watanabe, M., Ehime University, Matsuyama, Japan; Tanaka, H., National Research Institute of Far Seas Fisheries, Shimizu, Japan. The procellariformes such as albatross and petrel are collected from the northern North Pacific (NNP) and southern part of the Indian Oceans (SI) and their subcutaneous fat were employed for organochlorine (OC) analysis. Black-footed albatross from NNP showed the highest OC contamination, some of them accumulated with PCB and DDT levels more than 100 and 50 μg/g wet wt, among all species of birds, respectively. Among the birds form SI, northern giant petrels exhibited higher organochlorines contamination which could be attributable to their scavenging feeding habits. Yellow-nosed albatross found with the lowest organochlorines accumulation may be due to their feedings with lower trophic levels. Next to PCBs, DDTs showed higher concentrations followed by chlordanes, HCB and HCHs. IUPAC 153 had highest contribution to the total PCBs and IUPAC 126 was higher among non-ortho coplanar congeners in many samples. The calculated toxic equivalents (TEQs) for the PCBs showed that birds from NNP exhibited 1 order of magnitude higher TEQs than those from the SI, indicating that oceanic birds from northern hemisphere have higher exposure to these xenobiotics. The estimated hazard index (HI) suggested that the accumulation of toxic PCB congeners in some species of these remote oceanic birds exceeded the threshold levels of which most or least sensitive birds exhibited adverse effects, implying the possible toxic impacts by some PCB congeners on these populations.

345 Change Point Regression of Organochlorine Contaminants in Herring Gull Eggs (1974-1997) and the Relationship with Winter Severity. Pekarik, C.*, Canadian Wildlife Service, Downsview, ON, Canada; Hebert, C.E., Canadian Wildlife Service, Hull, PQ, Canada; Weseloh, D.V., Canadian Wildlife Service, Downsview, ON, Canada. Organochlorine contaminants in herring gull eggs were measured at 12 colonies on the Great Lakes. Log transformed data were analyzed using change point regression. The model incorporated three parameters: a large change in contaminant concentration (at the change point), and two slopes, one before and one after the change point. For PCBs, results indicated that the majority of the analyses (58%) showed two equal slopes before and after a significant change in PCB levels; 17% of the analyses showed two different slopes before and after a significant change in PCB levels; 17% of the analyses showed two different slopes, but no significant change in PCB levels at the change point; 8% of the analyses were best represented by a single trend. In order to explain significant increases or decreases at the change points, winter severity data were analyzed. When winter severity was incorporated into the change point models, the Niagara River was the only colony where it deemed the change in concentration at the change point non-significant. However, partial Pearson correlations (accounting for temporal changes) between PCB and winter severity indicated that winter severity tended to be significant at colonies in the upper Great Lakes. This would appear to indicate that winter severity has affected PCB levels in herring gull eggs, and that this effect occurs more often in colonies from the upper Great Lakes. Nonetheless, winter severity alone does not explain the largest year to year changes in temporal trends of organochlorine contaminants.

346 Biological effects of chlorinated hydrocarbon contaminants in osprey chicks. Elliott, J.E.*, Wilson, L.K., Canadian Wildlife Service, Delta, BC; Henny, C.J., U.S. Geological Survey, Corvallis, OR; Trudeau, S., Kennedy, S.W., Canadian Wildlife Service, Hull, QU; Leighton, F.A., University of Saskatchewan, Saskatoon, SK; Cheng, K.M., University of British Columbia, Vancouver, BC. During the 1995 and 1996 breeding seasons, eggs of ospreys (*Pandion haliaetus*) were collected within a gradient of chlorinated hydrocarbon exposure along the Fraser and Columbia River systems of British Columbia, Washington and Oregon. Fifty-one eggs were placed into a laboratory incubator, of which 39 hatched; chicks were sacrificed within 24 h. There were no differences in hatching success between treatment and reference areas; thus, low productivity of ospreys nesting downstream of the pulp mills at Kamloops and Castlegar is not caused by poor embryonic survival. Residual yolk sacs of eggs collected downstream of Castlgar, BC, contained greater concentrations of 2378-TCDD (mean = 3050 $\pm$ 990 ng/kg lw) compared to other sites (eg. 45 $\pm$ 36 ng TCDD/kg lw). Total PCBs in yolk sacs were also higher at that site and in samples from the Lower Columbia River near Portland compared, to reference areas. Hepatic EROD activity was greater (2.8-fold) in chicks collected downstream of Kamloops compared to a reference site, and correlated positively with lipid-normalized concentrations of total PCBs and CB-126, but not 2378-TCDD. A hepatic cytochrome P4501A (CYP1A) cross-reactive protein was detected in all samples tested, and correlated with EROD activity. Very low concentrations of retinol and retinol palmitate were found in all liver, kidney and plasma samples. Vitamin A was present mainly in the form of 3-dehydroretinol and two unknown esterified forms. Analysis of other morphological, physiological and histological parameters is underway.

347 Predicting Starling Chick Carcass Pcb Concentrations from Pcb Concentrations in Ingested Animal Matter. Arena, S.M.*, Southern Illinois University, Carbondale, IL; Halbrook, R.S., Southern Illinois University, Carbondale, IL; Arenal, C.A, Southern Illinois University, Carbondale, IL. Pre-remediation studies at Crab Orchard National Wildlife Refuge (CONWR) revealed statistically significant accumulations of PCBs and effects in starling chicks (*Sturnus vulgaris*) from contaminated sites. Although the expected route of exposure for starling chicks is thought to be via the diet, this has not previously been documented. The purpose of this study was to quantify material in the stomachs of chicks collected from contaminated sites on CONWR and to measure PCB concentrations in animal material present in the stomachs. During 1995 and 1996, 14 and 22 stomachs were removed from starling chicks collected from contaminated and reference locations, respectively. The percent dry weight of animal, plant, and mineral matter in stomach contents were determined and animal matter from individuals from the same nest were combined to form composite samples for PCB (Aroclor 1254) analysis. Stomach contents were composed mostly of animal matter (84%) and the percent animal matter did not differ significantly among locations. The mean PCB concentration in composite samples of animal matter from stomachs of chicks collected from contaminated sites (1.82± 1.17 ppm) was significantly greater than the concentrations in composite samples from reference locations ($\leq$ DL, 0.24 ppm). PCB concentrations in stomach contents were significantly correlated with concentrations in carcasses of chicks from which the stomachs had been collected. The equation $Y = 0.556 + 12.39 * X$ (Y = starling chick PCB concentration, X = stomach animal matter PCB concentration), is useful in predicting starling chick carcass PCB concentrations from their stomach PCB concentrations. These data suggest that a significant portion of the PCB body burden in starling chicks results from ingestion of PCB contaminated animal matter in the diet.

348 A Duel Label Method for Determining Assimilation and Toxicokinetics of 2,2',4,4',5,5' - Hexachlorobiphenyl in the Gut Tract of Ring Doves fed a Contaminated Diet. Drouillard, K.G.*, Trent University, Peterborough, ON, Canada, Norstrom, R.J. Canadian Wildlife Service, Hull, PQ, Canada. A gastro-intestinal fugacity gradient mechanism has been used to describe the phenomenon of biomagnification in fish. The application of such a model in the avian system was explored using the Ring Dove (*Streptopelia risoria*) fed a diet of pigeon pellets containing 2,2',4,4',5,5'-hexachlorobiphenyl (CB#153), a highly persistent and negligibly metabolized PCB congener. Ring doves were fed a diet containing CB#153 at a concentration of 120 ug/kg for 25 d and then switched to a new diet of 13C-CB#153 at the same concentration for a further 60 d. Replicate birds were sacrificed at 4 h, 8 h and 18 h after feeding the new diet and over geometric time intervals for the remainder of the experiment. Birds fed a single meal of 13C-CB#153, and sacrificed within 24 hours, exhibited declining trends in 13C-CB153 concentrations along the gut whereas unlabeled CB#153 concentrations exhibited negligible or slightly increasing trends. The maximum fugacity gradient observed between upper and lower gut sections was 2.90. A steady state BMF was calculated based on the difference in lipid contents between food and feces (8.3 fold) and the dry mass assimilation efficiency of diet (0.44) to yield a maximum BMF of 8.35 for CB 153 in this species. The calculated BMF was lower then field BMFs typically reported for other avian species on the order of 30. The different trends in concentration profiles between recently assimilated 13C-CB153 and native CB153 along various gut tract sections may indicate that contaminant fluxes associated with uptake during digestion and elimination to feces may be decoupled during a feeding event.

349 Derivation of Protein Expression Signatures of Five Pesticides Using Digital Image Analysis. Bradley, B. P.*; J. L. Shepard. University of Maryland (UMBC), Baltimore, MD. Northern Bobwhite Quail (*Colinus virginianus*) were exposed to five pesticides, methomyl, parathion, pentachlorophenol, 2,4 dinitrophenol and a proprietary compound (XQT). The goal was to isolate, from the entire protein profile, subsets of proteins (signatures) specific to each chemical. Soluble proteins were extracted from the cellular component of blood samples collected 24hr. after dosage with one of the chemicals. There were 20 birds per treatment, including controls dosed with solvent (corn oil) only. Proteins were separated using two-dimensional gel electrophoresis (2D-PAGE). The protein features on the 2D maps were scanned, digitized and composite images made by pairwise comparisons of images using MELANIE II™ software (Bio Rad). Composite images from each of the five chemicals were compared using MELANIE. Each chemical had a unique expression signature, consisting of proteins present or absent in the profile of that chemical compared to all others. Using similar methods, we compared protein profiles from toxic and nontoxic levels of compound XQT. Some proteins were present and some absent only in one concentration, some were present and some absent in both concentrations. Thus exposure and effect may be separable. Finally we have examined conservation of signal across taxa. We found a considerable common component of signal between quail and mallard ducks (*Anas platyrhynchos*). The next steps are to consider proteins varying in levels of expression and then identify the key proteins diagnostic of each chemical and concentration.

350 Evidence of Soil and Sediment Ingestion in Wild Birds and Mammals. Scanlon, P.F.*; Connor, E. E.; Crawford, J.A.; Russell, A.J.M.; Gibson, D.F., V.P.I. & S.U., Blacksburg, VA. Ingestion of soil and sediment presents a little recognized route by which environmental contaminants may be ingested. Such ingestion may be deliberate or be an inadvertent action resulting from ingestion of forage or prey. Means of estimation of soil or sediment ingestion using feces have been developed. Essentially these have utilized 2 approaches. Use of elements as tracers has considered elements which are not incorporated in plants, but are present in soils. One example is titanium (Ti). Another approach is to use acid insoluble residue (AIR) in feces and consider this as a measure of soil on sediment ingestion. Connor (1993, MS Thesis, VPI & SU) considered both approaches and showed that both had merits and advantages. This report concerns the estimation of soil ingestion as percentages of dry matter ingestion from feces of a range of species together with AIR's as Indices of soil ingestion for a further range of species where less was known about their food habits and digestibilities of food items. Estimates (mean± SE) of soil ingestion as percentage of dry matter intake were as follows: mallard (Anas platyrhynchos), 11.7± 1.5%; Canada goose (Branta canadensis), 4.9± 0.6; mourning dove (Zenaida macroura), 0.8± 0.4%); short-tailed shrew (Blarina brevicauda), 5.2± 1.9%; white-footed mice (Peromyscus leucopus), 16.2± 4.9; and meadow vole (Microtus pennsylvanicus), 2.0± 0.3%. Percentage of AIR's Residues in feces of Australian species exceeded 10% for Euros (Macropus robustus) from 4 locations in the Northern Territory, for feral donkeys (Equus asinus) in the Northern Territory, for Eastern Grey kangaroos (Macropus giganteus) in Victoria, and for rock wallabies (Petrogale sp.) in 2 locations in the Northern Territory. Agile wallabies (Macropus agilis) had lower AIR's (5-8%) at a location in Darwin, N.T. AIR values for North America and Irish deer species were lower than 5% as were those of European rabbits. Soil ingestion is substantial in a wide range of species and represents a route by which contaminants may be acquired which currently is little considered.

351 A Critique of Aquatic Life Criteria for Metals. Birge, W.J.*, Spromberg, J.A., Shaw, J.R., University of Kentucky, Lexington, KY. Under the Clean Water Act, the U.S. EPA developed acute and chronic criteria for the protection of aquatic life. Metal criteria are based on toxic effects of total recoverable or dissolved concentrations present in the water column. The exclusion in criterion development of alternative avenues of exposure, ecological factors affecting survivorship, and other phenomena (*e.g.* metal mixtures) compromised the integrity of the process and, therefore, there are no reliable estimates of numbers of species protected within the U.S. It appears likely that many species, due to habitat selection, conditions that enhance exposure, or characteristics that limit survivorship, fall outside the protective loop. Frequencies of species extinction or endangerment exceed prehistoric levels and undetectable species attrition rates of 0.2 to 0.5% per year could prove catastrophic within the next century. We intend to present ecological evidence that metal mixtures potentiate toxicity; that food web transfer of metals from a primary producer to a primary consumer, in the absence of water column exposure, limits or precludes reproduction; and that habitat selection, physiological characteristics (e.g. feeding, respiration), life cycle complexities and/or trophic dependencies may enhance metal exposure or limit survivorship in certain aquatic species (*e.g.* molluscs). Discussion will be given to the development of survivorship indices (SI) that may be integrated with toxicity data to developing more ecologically-based regulatory criteria. Protecting biodiversity and maintaining generational continuity over time will require attention to subtle degrees of impact that fall within the limits of natural variability observed in aquatic communities. Rectifying present deficiencies in existing regulatory strategies and meeting challenges of future decades will require full and objective dialog, as well as development of new technology and modeling concepts that integrate ecological, physiological, and toxicological perspectives.

352 Uses and Abuses of Metal Speciation in the Development of Alternative Water Quality Criteria. Bedsworth, W.W.; Sedlak, D.L.*, University of California, Berkeley, CA. Measurements of speciation in metal-polluted waters by techniques such as cathodic stripping voltammetry, chelating resin column partitioning and high performance liquid chromatography (HPLC) indicate that many pollutant metals are complexed by strong ligands. Previous studies have demonstrated that that these metal-organo complexes are less toxic to aquatic organisms than predicted by current water quality criteria. To account for metal speciation in water quality criteria, it is tempting to use equilibrium speciation models. However, improper application of these models can lead to water quality criteria that are not adequately protective of sensitive organisms. The behavior of the synthetic chelating agent ethylenediaminetetraacetate (EDTA) in wastewater effluent and in surface waters provides an example of the potential uses and abuses of equilibrium speciation models. Research conducted in our laboratory has shown that some metals pass through wastewater treatment plants as metal-EDTA complexes. Equilibrium calculations also indicate that soft metals, such as lead and cadmium, will be converted into more toxic metal-chloro complexes once the wastewater is discharged to seawater. Other metal-EDTA complexes, such as $CuEDTA^{2-}$ and $NiEDTA^{2-}$, are predicted to be stable in freshwater and seawater after discharge. Equilibrium models neither consider complexation kinetics nor do they describe all of the possible processes that occur in these complex systems. Therefore, field measurements are required to test the stability of the complexes in the presence of other ligands, mineral surfaces and biological processes before speciation models can be used to revise water quality criteria.

353 Uncertainties in Water Quality Criteria and Their Estimation Techniques: Tributyltin and Copper. Cardwell, R.D.*, DeForest, D., Cothern, K., Parametrix, Inc., Kirkland, Washington. This paper analyzes uncertainties in the laboratory data used to define concentration-effect relationships and saltwater water quality criteria for tributyltin and freshwater criteria for copper. We examined the magnitudes of uncertainties around concentrations estimated to protect 95%, 90% and 80% of the aquatic species, and differences arising from use of different estimation techniques. Data constituted acute toxicity tests comprising the U.S. EPA water quality databases for these two chemicals, namely individual LC50s, LC50s averaged by species, and LC50s averaged by genus. Four estimation/curve-fitting techniques were used: Stephan et al. (1985), Aldenberg and Slob (1993), logistic regression, and non-parametric (Gilbert 1987). Criteria estimates were insensitive to whether the data analyzed comprised LC50's, species mean acute values, or genus mean acute values. Some of the curve-fitting techniques materially influenced criterion magnitudes. Both the Aldenberg and Slob (1993) and logistic regression techniques produced criteria that were 42-61% (TBT) and 37-59% lower (Cu), respectively, than the Stephan et al. method. In contrast, the non-parametric technique produced criteria that were generally comparable to Stephan et al. (especially at the 5% protection level). Uncertainty was expressed as 95% confidence intervals around predicted criteria.

354 Selenium Water Quality Criteria: The Need For Revision. W. J. Adams, Kennecott Utah Copper, Magna, Utah, K. V. Brix and J. Toll, Parametrix, Inc., Kirkland, Washington. The acute (20μg/L) and chronic (5μg/L) water quality criterion (WQC) for selenium (freshwater) were established in 1987. In 1996, USEPA proposed to revise the freshwater acute criterion for the Great Lakes System using an additive model based on the relative forms of selenium including selenate, selenite and other reduced (organic) forms. The proposed revision of the acute criterion was in recognition of the need to address the various selenium forms and the need to update the database to reflect several additional studies. These needs still exist and EPA is revising the database. Recently, the US Fish and Wildlife Service has advocated that the chronic criterion be revised to 2.0 μg/L. This recommendation is based on field studies with birds derived primarily from the San Joaquin Valley, California. Should this recommendation be adopted the consequences are significant. The present paper reviews recent significant additions to the selenium literature including freshwater aquatic and avian studies performed by the authors, and discusses the importance of chemical speciation and site-specific considerations. Recently collected data on background selenium concentrations in surface waters in the western US will be reviewed in relation to the existing WQC (5 μg/L) and the proposed criterion (2 μg/L). The need to update the WQC for selenium is supported, but the data do not appear to support a national criterion of 2 ug/L. An alternative risk-based approach for revising the national chronic criterion which would allow the use of site-specific data is recommended.

355 The Selenium Aquatic Toxicity Model: A Framework for Defining Research Needs and Determining Water Quality Criteria. Carlton, R.G.*, Electric Power Research Institute, Palo Alto, CA; Bowie, G.L., Tetra Tech, Inc., Lafayette, CA; Porcella, D.B., Electric Power Research Institute, Palo Alto, CA. Selenium is a naturally occurring trace element which is essential for a variety of microbial, plant, and animal life forms. However, at elevated concentrations in the environment and in food selenium can be toxic. Recent research has identified significant differences in the relative toxicities and biogeochemical pathways and fates of the various forms of selenium [Se(IV), Se(VI), Se(0), Se(-II), Se-org]. The complexity of the biogeochemistry, bioaccumulation, trophic transfer, and toxicity of Se species has been parameterized in a mechanistic computer model (the Selenium Aquatic Toxicity Model) to calculate the ecological effects of Se and more accurately determine water quality criteria. Also, the model supplies a framework that helps focus experimental research and integrates results and data from the literature. The efficacy of the model will be demonstrated in the context of a case study of a hydrologically complex, Se-contaminated reservoir, for which the model favorably predicted Se dynamics, speciation, and long-term fate of Se loadings consistent with monitoring data.

356 Equilibrium Modeling of Metal-gill Interactions: a Framework for Better Understanding the Effects of Waterborne Metals on Aquatic Organisms. Playle, R.C. Wilfrid Laurier University, Waterloo, Ontario, N2L 3C5. Waterborne metals can bind to gills of freshwater fish and disrupt the ionoregulatory and respiratory functions of the gills. Competing cations such as Ca^{2+} and compelling ligand such as dissolved organic matter (DOM) reduce these effects by preventing metals from binding to the gills. The gill membrane can be considered as a compelling ligand, and conditional equilibrium stability constants can be calculated for the strength of metal binding to the gill membrane (log $_{Metal\text{-}gill}$). Once conditional stability constants are calculated they can be inserted into aquatic chemistry programs, producing powerful predictive tools which include metal concentration, metal speciation, competitive effects due to cations such as Ca^{2+}, and includes complexation by DOM, all of which affect metal binding to fish gills and therefore metal toxicity to fish. Traditional water chemistry modeling, or use of ion selective electrodes, do not take into account competitive effects or the physiology of biological membranes. For example, water chemistry modeling alone can not predict the greater protective effect of Ca against Cd toxicity compared to the effect of Ca against Ag toxicity to fish. This talk will be a critical review of the metal-gill binding approach, and its contributions to the understanding of the influences of water chemistry on the physiological and toxicological effects of metals towards fish and to other aquatic organisms.

357 Use of a Biotic Ligand Model to Calculate Site Specific Water Effect Ratios for Metals. Santore*, R., HydroQual Inc., Mahwah, NJ; McGrath, J., HydroQual Incorporated, Mahwah, NJ; Kevin Brix, Parametrix, Kirkland, WA; Paquin, P., HydroQual Incorporated, Mahwah, NJ; Di Toro, D. HydroQual Incorporated, Mahwah, NJ and Manhattan College, Bronx, NY. The USEPA allows site specific adjustments to water quality criteria for metals by determination of water effect ratios (WERs). The WER is calculated from side by side toxicity tests using a *downstream* water (i.e., upstream plus effluent mixed in proportion to the respective flows) and a reference laboratory water. The use of a WER allows consideration of site specific water chemistry on the bioavailability of a metal. The disadvantages of determining WERs are the cost associated with toxicity testing and the variability of test results which are often observed. We have developed a methodology for applying a biotic ligand model to predict the bioavailability of copper and other metals as a function of water chemistry. The model includes detailed speciation of the metal, plus interaction with a biotic ligand defined as the site of action of acute toxicity. For example, the gill is the biotic ligand for acute metal toxicity to freshwater fish. The model was calibrated using measurements of metal accumulation in fish gills as a function of water chemistry (ranges of values for pH, DOC, and hardness) and was tested against measured LC50 values as a function of water chemistry. The model is able to predict the effects of site specific water quality on metal LC50's, and has been used to calculate WERs. Example predictions will be presented showing favorable comparison with measured WERs for copper.

358 Validation Study of Gill Model for Copper and Silver Using Water Effect Ratios. Brix, K., Henderson, D. Parametrix, Inc., Kirkland, WA.; Paquin, P., Hydroqual, Inc., Mahwah, NJ.; Adams, W., Kennecott Utah Copper, Magna, UT.; Gorsuch, J., Eastman Kodak Company, Rochester, NY. Over the past several years, various researchers have been actively developing a model to predict metal bioavailability and subsequent toxicity. Conceptually, this model predicts metal bioavailability by treating the fish gill as a ligand within the water matrix. The gill competes with other ligands such as those associated with organic carbon to form metal complexes. In addition to organic ligand competition, the model takes into account metal metal speciation and partioning within the matrix based on information such as pH and total suspended solids. Accurate prediction of gill metal concentrations can then be directly related to acute toxicity. In effort to validate this model for copper and silver, we conducted a series of Water Effect Ratio studies on different effluent-receiving water combinations. Water quality data from these studies were then input to the gill model to determine if it could accurately predict the Water Effect Ratios which were observed. Water Effect Ratios were performed with *Ceriodaphnia dubia* and *Pimephales promelas* on several effluents and receiving waters with a variety of water quality characteristics. Results of the Water Effects Ratios and gill model predictions are presented.

359 Application of the Acute Gill Binding Model to Trout Chronically Exposed to Cadmium. Hollis, L.M.*, McMaster University, Hamilton, ON, Canada; McGeer, J.C., McMaster University, Hamilton, ON, Canada; Playle, R.C., Wilfrid Laurier University, Waterloo, ON, Canada; McDonald, D.G., McMaster University, Hamilton, ON, Canada; Wood, C.M., McMaster University, Hamilton, ON, Canada. An acute gill surface binding model, originally developed in soft water, has been used to predict metal-gill interactions of fish and has been shown to be indicative of toxicity. We examined the application of this acute model to juvenile rainbow trout chronically (30 d) exposed to sublethal levels of cadmium (3 and 10 μg/L) in moderately hard (140 ppm as CaO_3) water. Gill cadmium burdens of chronically exposed fish resulted in 20-40 fold increases in cadmium levels compared to those levels predicted by the gill binding model to cause mortality during acute exposure (3 h). The model was able to accurately predict acute cadmium accumulation in gills of control fish; however, high background accumulation of cadmium in gills of acclimated fish prevented detection of any further accumulation with an acute exposure. To increase the sensitivity of detected gill cadmium binding in acclimated fish, radioactive ^{109}Cd was used in an acute exposure. A small saturable binding component was observed in acclimated fish; however, this accumulation was not indicative of a toxic response. Consequently, the current acute gill binding model can be used to predict metal accumulation in naïve fish exposed to cadmium in hard water for acute exposures but it cannot be applied to trout that have been chronically exposed. This work was funded by an N.S.E.R.C. Strategic Program in Environmental Quality, The International Copper Association Ltd., Cominco Ltd., and Falconbridge Ltd.

360 a Comparison of the Intestinal Metal Bioavailability of Cu, Cd and Zn in Rainbow Trout (*Oncorhynchus mykiss*). Baskin, S.J.*, S.J. Clearwater, C.M Wood and D.G. McDonald. McMaster University, Hamilton, ON, Canada. Dietary bioavailability of copper, cadmium and zinc to 0.3 kg rainbow trout (*Oncorhynchus mykiss*) was examined at 15 °C at 1, 2, 3, or 7 days following a single bolus dose to the stomach of 0.5 μmol of radiolabelled metal. After exposure, fish were terminally anaesthetized and all internal organs and carcass were individually counted for radioactivity. Fish exposed to Zn had absorbed 20.0% of the dose by 24 hr. 21.0% was bound up in the gastrointestinal tissues and the remainder of the dose was either excreted (38.1%) or was present in the gut lumen (20.9%). Six days later, the amount absorbed had increased slightly to 30.5% with only 11.7% remaining in the gut (bound + luminal contents). The remainder was excreted. Cadmium showed a much different pattern of uptake. Only 3.8% of the dose was absorbed after 48 hr, with the remainder either in the gut (60.9%, bound + luminal) or excreted (35.3%). There was no further uptake of Cd. In fact, after 7 days, only 1.7% of the dose remained in the body with 57.7% being excreted. Cu absorption was intermediate to Zn and Cd at 11.7% after 3 days with 18.5% bound to gut tissues. It is concluded that metals are retained much longer in the gastrointestinal tract than food. Zinc is rapidly distributed to a large number of organs throughout the body, while Cd distribution is limited and is largely retained within the gut tissues. Internally the main target of Cu is the liver but a large portion also remains bound to the gastrointestinal tissues.

361 Induction of Testis-Ova in Japanese Medaka by Estrogen Agonists. Metcalfe, C.D.*, T.L. Metcalfe, M..A.. Gray and Y. Kiparissis, Trent University, Peterborough, ON, Canada; A.J. Niimi, Fisheries and Oceans, Burlington, ON, Canada. Recent observations of intersex characteristics in fish downstream of sewage treatment plants (STPs) in the U.K. have led to speculation that estrogenic compounds in STP effluents may be altering gonadal development in these fish. However, before this cause- and-affect linkage can be established, studies are needed to determine the factors that influence alterations to gonadal development in teleosts. Our studies have shown that exposure of Japanese medaka, *Oryzias latipes* over a period from hatch to 3-4 months of age to aqueous solutions of octylphenol, nonylphenol, nonylphenol mono- and diethoxylate, o,p'-DDT and ethinylestradiol induce development of testis-ova; an intersex condition characterized by both testicular and ovarian tissue in the gonad. Studies with the yeast estrogenicity (YES) assay indicate that all of these test compounds are estrogen agonists. Negative results were obtained in both the *in vitro* YES assay and *in vivo* medaka assay with diethylhexylphthalate (DEHP) and atrazine; two compounds with endocrine disrupting potential that do not appear to be estrogen agonists. Optimal induction of testis-ova occurs when medaka are exposed to estrogen agonists within 3 days post-hatch, but exposure of adult medaka can also induce this intersex condition at low incidences. Tests are currently underway to determine whether exposures of female medaka to o,p'-DDT can lead to transgenerational induction of testis-ova in offspring. These protocols with medaka may be a model test system for studies of alterations to gonadal development in fish.

362 Effects on Growth, Development and Fertility of the Natural Steroid Hormone 17β-estradiol in Fathead Minnow (*Pimephales promelas*). Lange, R., Schering AG, Berlin, Germany; Dionne, E., Springborn Laboratories, Wareham, MA. Fathead minnow (*Pimephales promelas*) were exposed to 17β-estradiol in order to evaluate the effects on growth and development in early life and juvenile stages as well as on fertility in mature adults. In the early life stage (ELS) study embryos and fish fry were exposed to 17β-estradiol at concentrations of 1, 2, 7, 22, 64, 170 and 560 ng/l and controls over 29 days post hatch. Subsequently, a subset of the exposed and control fish were continued in an extended early life stage test for 63 days (92 days posthatch) at concentrations of 1, 3, 8, 21 and 67 ng/l. Additionally, mature fish were exposed in breeding pairs over 42 days at concentrations of 15, 30, 60, 120 and 240 ng/l and controls. Test concentrations were analytically monitored by an HPLC/UV detection method. In the ELS study hatching success was affected at the highest concentration as well as survival. A marked effect on growth (weight increase reduction) was observed after 28 days at the concentration of 170 ng/l. The EC50 for length increase was 270 ng/l. In the continued ELS exposure survival was effected at 67 ng/l. Length and weight increase was affected at concentrations of 8 ng/l and higher. The EC50 was 29 ng/I for weight increase. At the two highest concentrations (21 and 67 ng/I) the fish showed marked deformities including an enlarged ovipositor in some fish and a tadpole-like appearance. At 92 days posthatch some of the fish showed already secondary sex characteristics. In the controls and the lowest concentration male and female fish were observed. In the higher concentration no male fish was found. In the study with mature adult fish the fertility was not affected at a statistically significant degree in the exposure groups. The study indicated that the natural steroid hormone 17β-estradiol, which was detected in some surface water samples in the UK, has marked effects on growth and development of fish. The early life stages of fish seemed to be more sensitive to hormonal effects than the adults. The developmental effects could be detected by growth measurements. The feminization of the fish under exposure to 17β-estradiol is a likely effect. Histopathology is in progress in order to evaluate the effects on sex determination.

363 Compensatory Mechanisms for Maintenance of Fish Populations in Contaminated Freshwater Streams. Greeley, M. S., Jr.*, Adams, S. M., Peterson, M. J., Ryon M. G., and Southworth, G. R., Environmental Sciences Division, Oak Ridge National Laboratory, TN. Redbreast sunfish, *Lepomis auratus*, survive and often thrive in contaminated streams lacking populations of more pollution-sensitive fish species. Compensatory mechanisms that could account for the relative insensitivity of sunfish to aqueous contamination were examined in populations inhabiting three streams on the DOE Oak Ridge Reservation. Each stream is affected by drainage and wastewater discharges from one of three DOE facilities on the reservation: East Fork Poplar Creek by the Y-12 Plant, White Oak Creek by Oak Ridge National Laboratory, and Mitchell Branch by the East Tennessee Technological Park (formerly the K-25 Site). In over a decade of biomonitoring activities, bioaccumulation of various contaminants, especially mercury and other divalent metals and organics such as polychlorinated biphenyls (PCBs), has been well-documented in sunfish from these streams. Molecular, biochemical and physiological responses typical of contaminant-stressed populations have been observed. And survival of redbreast sunfish embryos has been shown to be adversely affected by exposure to water from the streams in acute laboratory toxicity tests. However, redbreast sunfish populations persist in each stream, albeit often in association with abnormal population characteristics. Redbreast sunfish appear to compensate for contaminant stress in these streams through various reproductive mechanisms. One is a reduction in age-at-maturity, which has been especially pronounced in female sunfish inhabiting the upstream reaches of East Fork Poplar Creek. Another is increased fecundity, which has been observed at downstream locations in all three streams, especially Mitchell Branch. As these streams have recovered in response to remedial actions, these reproductive characteristics have trended back towards normality.

364 Effects of Paper Mill Effluents on Reproduction and Health of Largemouth Bass (*Micropterus salmoides*) in Florida. Sepúlveda, M. S. *, Gallagher, E., Shoeb, T., Denslow, N.D., University of Florida, Gainesville, FL; Holm, S., Georgia-Pacific Corp., Atlanta GA; Gross, D.A., Ruessler, S. and Gross, T. S., USGS-BRD. Gainesville, FL. Previous results from our laboratories have indicated altered endocrine parameters for largemouth bass at the St. John's River - Rice Creek confluence. These alterations, including decreased systemic gonadal steroids and vitellogenin, may be related to local exposure to chemical components within papermill effluents. Therefore, a multi-exposure experiment was conducted using large mouth bass to examine the effects of paper mill effluents on reproduction and health. Five hundred reproductively active bass were exposed to paper mill effluents in an outdoor tank facility. Fish were randomly assigned to one of 10 treatments (50 fish per treatment tank; 25 male and 25 female): 0%, 10%, 20%, 40%, and 80% effluent for 28 or 56-day exposures. Tanks were checked daily and water quality monitored bi-weekly. At the end of each exposure point fish were removed from each tank, weighed, measured, blood collected, and each animal sacrificed for a full health assessment. The following parameters were used to evaluate effects on reproduction: sex steroid/hormone concentrations, vitellogenin, gonadal somatic indices (GSI), gonad histology, and size/number of eggs in females. General health was assessed using several blood parameters and complete necropsies. Organ somatic indices, organ histology, and liver EROD activity were also determined. In general, health status/assessment was negatively impacted by increasing effluent dose and exposure length. Gonadal weights and GSI decreased with increased effluent dose and exposure length. These data suggest significant health and reproductive effects in largemouth bass exposed to papermill effluents at 20% or greater concentrations, which are levels normally occurring in effluent discharge sites.

365 Individual and Population Responses of the Gray-tailed Vole, *Microtus Canicaudus*, to Endocrine Disruptors. Wolff, J.O.*, Caslin, T., and Edge, W.D., Department of Fisheries and Wildlife, Oregon State University, Corvallis, OR. We conducted a field experiment to determine if a nominal application rate of commercial formulation of vinclozolin, a fungicide, which is a purported anti-androgenic endocrine disruptor, adversely affected reproductive development and demography of gray-tailed voles. We monitored demography in four control (water applied) and four treatment (vinclozolin applied) populations of voles in 0.2 ha enclosures. The experiment was conducted twice, once in tall grass during the rainy season, and once in short grass during the dry season. Representative male voles that were exposed to the treatment while they were in utero were later removed from the enclosures and examined for reproductive development. No statistically significant differences were found for testes size, seminal vesicle length, or reproductive competence in either study, but all measurements were smaller in treatment than in control animals. Similarly, we found no differences in population size, growth rate, juvenile recruitment, or sex ratio between voles in treatment and control populations. We conclude that a one-time standard application rate of vinclozolin does not adversely affect voles in their natural habitats, but we caution that repeated application or extensive use may have adverse effects on vole reproduction, development, and demography. In summer 1998, we are conducting a similar experiment following this same experimental design and protocol only using methoxychlor, an estrogenic insecticide. The results from that experiment will also be presented.

366 Endocrine Disruption and Developmental Toxicity in Daphnids. LeBlanc, G.A., Department of Toxicology, North Carolina State University, Raleigh, NC. Recent studies have shown that daphnids are capable of metabolizing testosterone to a variety of conjugated, hydroxylated, and oxido-reduced derivatives. Some metabolites are preferentially eliminated from the daphnids (i.e. glucose conjugates), while others are preferentially retained by the daphnids (i.e. oxido/reduced derivatives). In the present study, we demonstrate that exposure to certain chemicals (i.e. piperonyl butoxide, propiconazole, 4-nonylphenol) elicits changes in testosterone biotransformation characterized by a reduction in the production of glucose conjugated testosterone with a commensurate increase in the production of oxido/reduced derivatives. We refer to this effect as metabolic androgenization since these metabolic changes result in a decrease in androgen elimination and an increase in androgen accumulation. Chemicals that caused metabolic androgenization also interfered with the prenatal development of daphnids in a manner indicative of developmental retardation. Direct exposure of daphnids to the steroidal androgens testosterone and androstenedione, but not the estrogen diethylstilbestrol or the anti-androgen cyproterone acetate, also resulted in developmental retardation. These results suggest that metabolic androgenization is the mechanism by which the chemicals elicit developmental toxicity. Studies are underway to determine whether steroidal androgens elicit developmental toxicity by interfering with the provision of nutrients to the developing embryo.

367 Low Doses of Atrazine Increase Male Production in Daphnia pulicaria. Dodson SI*, Merritt CM, Shannahan J-P, Schults CM; Department of Zoology -- Birge Hall, University of Wisconsin, 530 Lincoln Drive, Madison, WI 53706-1381. We have developed a 6-day bioassay that uses the sex ratio of Daphnia pulicaria neonates as an endpoint. D. pulicaria is a practical indicator species because of its short generation time, wide distribution, and ecological importance. Previous tests with known chemicals (nonylphenol, dieldrin) suggest that sex ratio may be more sensitive than standard endpoints (fecundity or survivorship) as a measure of environmental health. Atrazine is a widely-used chemical that is of known health concern for aquatic organisms at concentrations above about 100 ppb. It occurs in the aquatic environment at concentrations mostly in the range of 0.1 to 5 ppb, and the EPA have set safe drinking water limits for life time human exposure at 3 ppb of atrazine. We performed about 25 bioassays in which we exposed Daphnia to atrazine concentrations in the range of 0.01 to 500 ppb. Atrazine had no effect on survival or fecundity of the adults, but it enhanced male production (increased sex ratio) at concentrations as low as 0.5 ppb. These results imply that the sex ratio endpoint is more than two orders of magnitude more sensitive to atrazine than are the survival or fecundity endpoints.

368 Effects of the Synthetic Oestrogen, Diethylstilbestrol, on the Survival, Development and Reproduction of a Marine Copepod (*Tisbe battagliai*). Hutchinson, T.H., Pounds, N.A., Hampel, M. & Williams, T.D.. Brixham Environmental Laboratory, ZENECA Limited, Freshwater Quarry, Brixham, Devon TQ5 8BA, United Kingdom. In order to conduct environmental risk assessments for potential endocrine disrupters, it is essential to address potential effects in invertebrate populations. Given recent reports suggesting that natural and synthetic oestrogens may be present in sewage effluent at levels which may impact on fish, it is pertinent to extend the hazard evaluation for such substances to aquatic invertebrates. Studies have therefore been undertaken to address the developmental and reproductive effects of diethylstilbestrol (DES) in *Tisbe battagliai* (Crustacea: Copepoda). This species was primarily selected in view of its utility as a test species for chronic bioassays, together with recent reports of abnormal (intersex) copepods sampled from coastal sites receiving discharges of sewage sludge. Copepod nauplii (<24 hrs old) were exposed to 100, 10, 1.0 and 0.1 μg DES per litre (nominal values) and effects first monitored in terms of survival, development and sex ratio at maturity. After 10 days exposure, 100 μg/l caused 100% mortality, with 0.1 - 10 μg/l having no impact on copepod survival. Examination of the animals at 10 days (adult stage in controls) indicated no effect on the F_0 sex ratio. Mature males and females were then paired and exposures continued to address reproductive output (total exposure period 21 days at $20 \pm 1\,^{\circ}$C). No further significant effects were observed for survival, development and reproduction (F_1 nauplii numbers), suggesting 21 day NOEC and LOEC values of 10 and 100 μg/l, respectively. The methodology and results of these studies will be presented and discussed with respect to the development of invertebrate test protocols for endocrine disrupting chemicals.

369 Biological Measures of Tributyltin Contamination: Is There Sex After Imposex? Evans, S.M.*, Dove Marine Laboratory, United Kingdom. Tributyltin (TBT) contamination causes the condition known as imposex in the dogwhelk, *Nucella lapillus*. Male genitalia, including a penis and vas deferens, become superimposed on the reproductive system of female dogwhelks. In severe cases, the condition can cause sterility, or premature death. Imposex in this species is evidently a specific response to TBT and has been developed as a biological indicator of TBT pollution. It has been used widely in monitoring the effectiveness of regulations restricting the use of TBT-based antifoulants to shipping vessels >25m in length. The paper will show how the condition can be used to identify sources of contamination and, by sampling populations of *N. lapillus* along gradients from them, the range over which there is a biological impact. It will consider current levels of contamination in: (i) areas adjacent to fish farms and marinas in countries in northern Europe where the use of TBT-based antifoulants is regulated and in Israel (using related whelks as biological indicators) where there is no regulation of their use; (ii) ports, harbours, and dry docks; and (iii) open seas. There will also be a discussion of the misuse of imposex as a biological indicator in some studies of TBT contamination.

370 Effects of Embryonic Pcb Exposure on Reproductive Development and Maturation in Japanese Quail *(Coturnix japonica)*. Henry, P.F.P.*, USGS Patuxent Wildlife Research Center, Laurel, MD; Casey, C.S., University of Maryland, College Park, MD; McGary, S., University of Maryland, College Park, MD; Ottinger, M.A., University of Maryland, College Park, MD. A multi generational study is in progress to evaluate potential effects of embryonic exposure to estrogenic PCB in the hypothalamic-pituitary-gonadal axis of the Japanese quail model. P1, F1 and F2 eggs are injected with a hydroxylated form of an estrogenic PCB congener (2',4',6'-trichloro-4-biphenylol) at day 4 of incubation. A number of variables are being analyzed in subsets of both male and female birds collected at three stages in the life cycle: pre-hatch, onset of sexual maturation, and during egg laying. Standardized measures of reproductive success such as % hatch, % 14 day chick survival are also recorded. Behavioral trials conducted at 21 days and at sexual maturation quantify relative activity (males and females) and readiness and effectiveness of courtship (males only). Results show that PCB-treated P1 males exhibited crowing and mounting behavior earlier than controls and that at 3 weeks, their non-injected offspring had variable but more elevated plasma testosterone levels than those measured in offspring of control birds. Because this PCB congener is known to be estrogenic in other animal model systems, present findings may counter assumptions that endocrine disruption occurs only via effects of steroid receptors. The initial data demonstrated earlier maturation among treated males; therefore endocrine disruption may be acting on neurotransmitter systems that later regulate GnRH and behavior. These neuroendocrine systems are evaluated in the PCB injected F1 and F2 offspring.

371 Uv Attenuation in Lake Superior. Vodacek, A.*, University of Maryland, College Park, MD; Green, S.A., Michigan Technological University, Houghton, MI. The penetration of UV solar irradiance into Lake Superior is highly dependent on local physical processes such as stratification, the thermal bar, and river plumes. We determined UV and visible attenuation across this range of water types in the spring and summer during two cruises along the Keweenaw Peninsula. We employed a Satlantic SeaWiFS profiling Multichannel Radiometer, with four additional UV channels between 300 and 400 nm, to measure downwelling irradiance as a function of depth. Attenuation in the UV was dominated by absorption by dissolved organic matter at all stations except those influenced by the high sediment load delivered to the lake by the Ontonogon River. Algorithms for estimating dissolved organic matter absorption in the lake from remote sensing data should be successful if the effects of a continental atmosphere can be accounted for in a regional atmospheric correction procedure.

372 The Role of Solar Photochemical Degradation in Regulating the UV Transparency and Spectral Characteristics of the Surface Waters of Lakes. Morris, D.P., and Hargreaves, B.R., Lehigh University, Bethlehem, PA. The role of photochemical degradation of dissolved organic carbon (DOC) on UV transparency (280-400 nm) has been investigated in several lakes of the Pocono Plateau. Diffuse attenuation coefficients ($K_{d\,UV}$) in the epilimnia of these lakes varied seasonally (39-81% decline from maximum $K_{d\,320\,nm}$), with minimum $K_{d\,UV}$ values occurring near summer solstice. Declines in $K_{d\,UV}$ corresponded to reductions in UV absorbance by dissolved material ($a_{d\,UV}$), caused by declines in both DOC concentration and UV absorptivity (a_d:[DOC]). The seasonal decline in $K_{d\,UV}$ was also accompanied by substantial increases in an absorption coefficient ratio ($a_{d\,250\,nm}$:$a_{d\,365\,nm}$) and decreases in spectral slopes (S). Experimental studies of photochemical degradation were also performed using filtered lake water and natural sunlight. Exposure to solar radiation produced a number of changes in optical parameters and DOC which resembled those observed in the water column: 1) $a_{d\,UV}$ (-35% to -52%), 2) UV absorptivity (-31% to -48%), 3) $a_{d\,250\,nm}$:$a_{d\,365\,nm}$ (0% to +39%), 4) spectral slope, S, (0 to -27%), and 5) DOC concentration (0% to -20%). Changes in $a_{d\,UV}$ were correlated with absorbed UV dose. The derived rate constants were used in a model to estimate the contribution of photochemical degradation of DOC to water column declines in $K_{d\,UV}$. Results suggest that rates of photochemical degradation were sufficient to account for the summer reductions in $K_{d\,UV}$ observed in the lakes.

373 Chromophoric Dissolved Organic Matter on the Continental Shelf off the Southeastern United States: its Optical Signature and Influence on Bio-optical Properties. Nelson, J.R., Skidaway Institute of Oceanography, Savannah, GA. The continental shelf of the South Atlantic Bight (SAB) extends some 120 km from the coast off Georgia. Bio-optical properties on this broad shelf can be strongly influenced by chromophoric dissolved organic matter (CDOM) which is discharged at the coast through a number of river estuaries. While a coastal frontal zone often restricts low salinity, high CDOM water to an inshore band, the frontal barrier can relax during periods of offshore wind stress, particularly when combined with relatively high fresh water discharge. Coastal waters can then be transported well offshore in a low salinity surface layer. The formation and dissipation of such features is strongly influenced by regional scale shifts in wind speed and direction. CDOM provides an optical indicator of these significant cross-shelf biogeochemical exchange events. During two periods of high freshwater discharge (Spring 1993 and Spring 1998), CDOM dominated total absorption (particulate + dissolved) from the UV well into the visible spectral region. Some 80% to >90% of the total absorption in the blue region was contributed by CDOM over a cross-shelf distance of some 50 km. With residence times of 1-2 months for water masses in the mid-shelf region, significant optical effects and interactions between UV and visible radiation and CDOM appear likely. Bio-optical and biogeochemical consequences of this include: a strong influence on remotely sensed reflectance; reduction in light penetration to depth; reduced primary production in the lower portion of the water column and at the sea floor; production of biologically available photo-products; and temporal changes in CDOM absorption and fluorescence properties in surface waters as chromophoric material bleaches.

374 The Effect of Dissolved Organic Matter on the Photolysis of Synthetic Organic Compounds in Antarctic Surface Waters. Miller, P.* and Chin, Y., The Ohio State University, Columbus, OH; McKnight, D., University of Colorado, Boulder, CO. The distribution and transformation of synthetic organic compounds (SOCs) in the Antarctic environment are poorly understood. The objective of this research was to determine if photochemical reactions mediated by dissolved natural organic matter played an important role in controlling the fate of SOCs in the surface waters of this extreme environment. Solutions of a "model" SOC, hexachlorobenzene, were prepared with either MilliQ water (as a control) or various Antarctic surface waters (i.e., Lake Bonney, Lake Hoare, and Lake Fryxell, McMurdo Sound, and Pony Lake) and exposed to the sunlight at McMurdo Station, Ross Island (77° 51') in quartz tubes. Light intensity for a given day was measured with chemical actinometry. Changes in hexachlorobenzene concentration were monitored throughout the course of an experiment via injections of hexane extracts onto a gas chromatograph equipped with electron capture detection. UV-VIS absorption spectra showed widely differing light absorption properties for the various waters. Pony Lake water had the highest amount of dissolved organic carbon (DOC) and attenuation in the solar region, and the lowest measured half-life (29 hours) of all samples. Waters having very low or no DOC (and little attenuation of natural light) did not have appreciable, measured half-lives. Thus, dissolved organic matter may be important in promoting the photochemical transformation of hexachlorobenzene in these waters.

375 Degradation of Halogenated Pesticides in the Presence of Nitrate and NOM in Aqueous Solutions Irradiated by Simulated Sunlight. Frimmel, F.H.*; Schindelin, A.J., Engler-Bunte-Institut, University of Karlsruhe, Germany. By means of simulated sunlight, the influence of NOM on the photochemical degradation of halogenated pesticides in the absence and presence of nitrate as precursor of the very reactive OH radicals in aqueous solutions and freshwater was investigated. Solutions of terbutylazine, dichlorprop, or chloridazon (a) in phosphate-buffered (pH = 8) demineralized water containing 50 mg/L nitrate or (b) in natural freshwaters were irradiated by a 1000-W Xe short arc lamp. The collimated beam was filtered using a combination of optical filters (WG 320 and WG 295) to fit the solar spectrum under summer midday conditions. In the absence of nitrate and NOM, the pesticides were degraded photolytically by simulated sunlight. The degradation rate constants were dependent on the absorption spectrum in the UVB range and the quantum yield of the degradation. As nitrate forms OH radicals by absorbing sunlight, the photochemical degradation of the pesticides was faster in the presence of nitrate. In the absence of nitrate, low concentrations of NOM accelerated the degradation due to the formation of reactive species by NOM. At higher concentrations of NOM, the inner filter effect of NOM lowered the degradation rate constants. In the presence of 50 mg/L nitrate, NOM decreased the degradation rate. In natural waters used for the experiments (nitrate concentrations between 2 mg/L and 15 mg/L), NOM acted mainly as light filter and as scavenger for the OH radicals. As a consequence, in most freshwater systems the accelerating effect of NOM by the formation of reactive species is of minor importance compared to the inner filter effect and radical scavenging.

376 Photochemically Enhanced Microbial Degradability of Dissolved Organic Matter in Lakes. Tranvik, L.J.*, Linkoping University, Linkoping, Sweden; Bertilsson, S., Linkoping University, Linkoping, Sweden. Numerous studies published in recent years emphasize the role of solar radiation in the degradation of dissolved organic matter (DOM) in lakes as well as in marine waters. Some of the DOM may become completely mineralized through photochemical degradation. In addition, the photochemical degradation may act in concert with the activity of heterotrophic microorganisms. In this way, recalcitrant DOM is photochemically degraded into labile organic intermediates, that are readily utilized by bacteria. We present evidence from northern temperate lakes for a major role of photochemically produced low-molecular-weight carboxylic acids as substrates for bacterioplankton production. In addition, we present a survey of water samples from 36 Swedish lakes, which were subject to standardized experiments on photochemical and microbial degradation. The lakes were selected to represent a wide range of environmental conditions, e. g. nutrient and humic concentrations, with the aim to assess which factors influence the potential role of the direct photochemical degradation of DOM, as well as the sequential photochemical-microbial degradation pathway.

377 Solar Radiation Induced Transformations of Dissolved and Particulate Organic Matter - Implications for Bacterial Growth and Carbon Cycling. Granéli, W.*, Anesio, A. M., Lindell, M., Lund University, Lund, Sweden; Tranvik, L., Bertilsson, S., Tema Vatten, Linköping University, Linköping, Sweden. We have investigated abiotic, solar radiation mediated mineralization of dissolved organic matter from lakes of varying humic content in Sweden and Brazil. We also studied photo oxidation of aquatic macrophyte detritus and leachate from macrophytes. Bacterial carrying capacity on photo transformed, dissolved (including surface and deep water from the Antarctic ocean) or particulate organic matter was quantified through bacterial batch cultures. Sterilised DOM or POM was exposed to natural or simulated solar radiation in quartz tubes and changes in DOC/POC, DIC, fluorescence and absorbance were measured. Abiotic, photo oxidative DIC production from DOM and POM was shown to be of a magnitude making this process important in the mineralization of dissolved humic matter and plant detritus in lakes. The rate of photomineralization for DOM was proportional to DOC content of the water. DIC production, loss of fluorescence and decrease in absorbance were well correlated. Most of the photo oxidation was due to the UV-A and PAR wavelength bands. Exposure of dissolved humic matter from lakes to solar radiation generally stimulated bacterial growth due to photo production of LMW organic molecules. However, no such effect was seen for water from the Antarctic ocean. For photoexposed macrophyte detritus or leachate there was also no growth stimulating effect or even a repression of bacterial growth.

378 UV Light Effects on Dissolved Organic Carbon (DOC) from a Prairie Wetland and Saline Lake. Waiser, M.J.* and R.D. Robarts, National Water Research Institute, Saskatoon, SK., Canada. Wetlands and saline lakes across the Canadian prairies contain some of the highest concentrations of DOC ever recorded. DOC is a potentially important carbon and energy source for bacteria in aquatic systems and UV light could play a major role in the transformation of refractory DOC to more microbially labile forms. The objective of this study was to establish if photolysis of DOC was occurring in a prairie wetland and saline lake and what impact this had not only on the bacterial community, but also on the structure and availability of the DOC itself. For photolysis experiments, filter sterilized water from the lake and pond was exposed to various spectra of natural sunlight. Post exposure, water was inoculated with the natural bacterial community and changes in bacterial numbers and production (^{3}H thymidine) were monitored. DOC was isolated from each system using XAD-8 resins and subjected to NMR, mass electrospray, and δ^{13}C analysis and ^{14}C dating. MW spectrum of the DOC was determined with a tangential flow cartridge (1000 D cutoff). Our studies indicate that photolyis occurred in both systems and had positive and negative effects on bacteria. Bacterial nutrient limitation played a strong role in determining whether or not bacteria ultimately utilized photolytically generated LMW carbon compounds. Photolytic processes appeared responsible for the low % of aromatics and high % of LMW DOC. In hydrologically closed basins, such as those studied, although a small % of this LMW carbon cycles quickly, we found that the majority was unavailable for microbial uptake. In some systems, photolytic breakdown of DOC over long time scales may result in a large pool of LMW compounds which are not biologically available.

379 Photoreactivity of Marine Dissolved Organic Matter: Differential Response of Bacterioplankton for Surface versus Mesopelagic Waters. Herndl, G.J.*, Obernosterer, I. Netherlands Institute for Sea Research, Texel, The Netherlands. During cruises in the Mediterranean and in the Caribbean Sea, surface and mesopelagic waters were exposed to surface solar radiation around noon for 6 h and subsequently inoculated with bacterioplankton from the respective sampling depths in order to determine the availability of the photoreactivated dissolved organic matter (DOM) for bacterioplankton. Bacterial activity measured via thymidine and leucine incorporation was 2 to 4 times higher in mesopelagic waters exposed to solar radiation as compared to treatments held in the dark. Surface waters collected from the chlorophyll maximum layer and exposed to solar radiation, however, supported significantly lower bacterial activity than water held in the dark. Additions of protein (bovine serum albumin, BSA) to the mesopelagic waters prior to exposure to solar radiation resulted in higher bacterial activity in the dark treatment, thus reversing the effect of solar radiation on the bioavailability of mesopelagic DOM. This tendency was not caused by an inability of bacterioplankton to use BSA since their activity in the BSA amended dark treatment was significantly higher than in the unamended treatments. Based on our experiments we conclude that, upon solar radiation, originally labile DOM becomes less available for bacterioplankton, while originally refractory DOM becomes more bioavailable for bacterioplankton.

380 Natural Photodegradation of Humic Substances and Humic-enzyme Complexes: Release of Fatty Acids and Enzymes. Wetzel, R. G.*, University of Alabama, Tuscaloosa, AL. Types and amounts of organic compounds were determined as they were generated by partial photolysis with natural UV-B or UV-A of dissolved humic substances (whole leachate; humic and fulvic acid fractions) released from different plant sources (emergent aquatic plants, floodplain shrubs, leaves of terrestrial trees) during decomposition under controlled conditions over one year. Solid state ^{13}C NMR indicated small changes to the macromolecules with increases in alkyl, carboxyl C and alterations of methoxyl C of lignin. GC-MS thermolysis indicated oxidative degradation of lignin with declines in cinnamyl phenols and increased derivatives of all lignin phenol families. HPLC analyses of side-chain oxidations of humic molecules showed photolytic release of large amounts of fatty acids (acetic, formic, citric, levulinic, pyruvic, tartaric, others). Differences in the efficacy of photodegradation occurred among DOM from different plant sources during the course of long-term partial bacterial decomposition. Photodegradative generation of fatty acids was generally greater among the more recalcitrant DOM after long periods of decomposition. Bacterial protein productivity increased directly in proportion to the differential photodegradative release of fatty acids. Dissolved humic substances from plant sources also formed complexes with phosphatases of bacterial and of algal origin and reduced hydrolytic activity by non-competitive inhibition. Restoration of hydrolytic enzyme activities from the humic substances-enzyme complexes increased progressively over time when exposed experimentally to natural and artificial UV irradiance. These data support the hypothesis that phosphatases and other enzymes can complex with humic substances that dominate the DOM pool by which the enzyme is temporarily inactivated, can be transported with water movements, and be displaced to other sites within the ecosystem. Upon exposure to UV irradiance in the photic zone, functional enzymes can be released.

381 Report of Progress on Understanding of "Contaminant-soil Interactions" from SETAC Pellston Conference, September 23-27, 1998. M. B. Tomson* and A. T. Kan, Department of Environmental Science and Engineering, Rice University, Houston, TX 77005 A conference entitled, *Assessing Contaminated Soils: From Soil-Contaminant Interactions to Ecosystem Management,* was organized by SETAC to assess the state of knowledge and research needs related to contaminated soils. A significant fraction of contaminants are commonly associated with the soil phase. Remediation is typically observed to be bimodal, with a large fraction of the adsorbed contaminant readily removable via normally understood mechanisms and with a second recalcitrant phase that resists desorption. These observations of resistant remediation are common for both organic chemicals and for inorganic compounds. Experts on the mechanisms of adsorption and desorption were invited to discuss the state of knowledge and research needs to address this important topic. The presentation will attempt to summarize the central thinking and the differences of opinion and suggestions of prioritized research needs and how these results might facilitate understanding contaminant-soil interactions.

382 Chemical Measures of Bioavailability. Alexander, M., Cornell University, Ithaca, NY. Considerable evidence exists that current chemical methods for analyzing soils often overestimate, sometimes by a considerable amount, the quantity of contaminants that are available to animals, plants, and microorganisms. Because bioassays are often slow, lack precision, and may not be accepted by regulators, a need exists for chemical procedures that show the concentrations that are available for uptake or are toxic to living organisms. This presentation will review methods for chemical measurement of bioavailability, properties of soil that may affect the results of these assays, characteristics of contaminants that may be important in designing a chemical assay, and possible difficulties associated with contaminant mixtures.

383 Mechanisms Limiting PAH Bioavailability in Soil. Roper, J.C.*; Pravecek, T.L.; Vanderford, M. and Pfaender, F.K University of North Carolina at Chapel Hill, NC. Determination of the bioavailability of a pollutant in a particular matrix is essential to risk assessment and ultimately to the establishment of remediation criteria. Polycyclic aromatic hydrocarbon (PAH) bioavailability is influenced by a number of factors including biotic transformations, abiotic transformations, aging, and the redox state of the soil. Laboratory studies in biotic systems have shown partial mineralization (14-45%) of fresh additions of ^{14}C labeled phenanthrene, pyrene and chrysene. After 270 days incubation, compounds are partially transformed and associated with the soil matrix (20%-35%). At the end of the incubation, no parent phenanthrene or pyrene was found in dichloromethane (DCM) extracts of soil. For chrysene, approximately 15% of the parent compound and 20% of the added label were in DCM extracts. Abiotic systems showed decreased total PAH extractability, but all the label recovered was associated with parent PAH. The behavior of freshly contaminated systems appeared to be very different from systems where contamination has been present for decades. Aged contaminated soils have significant quantities of extractable phenanthrene, pyrene, and chrysene which are persistent. This contamination has been shown to be partially available for biodegradation or association with the soil matrix under a variety of redox conditions during subsequent laboratory incubation. The persistence of PAHs in soils with an active and adapted degrader community may be explained by mechanisms of entrainment, sorption, and/or chemisorption. Each of these mechanisms dominate under various environmental conditions and have decidedly different ramifications for site risk assessment. Presently, these mechanisms are poorly distinguished by current laboratory techniques.

384 Chemical Methods to Estimate Bioavailable Arsenic in Contaminated Soils and Solid Media. Rodriguez, R.R.*, Sverdrup Environmental, Inc., St. Louis, MO; Basta, N.T., Oklahoma State University, Stillwater, OK; Casteel, S.W. and L.W. Pace, University of Missouri, Columbia, MO. The ability of chemical methods to evaluate bioavailability of arsenic in contaminated soil and solid media was determined. Chemical methods were an *in-vitro* method (IVG), that simulated the human gastro-intestinal environment, and soil chemical fractionation methods that extract different pools of soil arsenic.. Fifteen contaminated soils and solid media ranging from 401 to 17,456 mg As kg^{-1} were analyzed by IVG and fractionation methods. Arsenic measured by IVG and fractionation methods was compared with *in-vivo* bioavailable arsenic determined from feeding trials using immature swine. Arsenic extracted by the IVG stomach phase and intestinal phase were linearly correlated ($r^2 = 0.69$ and 0.67, respectively) with *in-vivo* bioavailable arsenic ($P < 0.01$). Analysis of variance showed the IVG stomach phase and intestinal phase were not statistically different from the *in-vivo* method. Five chemical extractants, ranging from deionized water to very aggressive reagents that dissolve occluded arsenic, were used to fractionate soil arsenic. The fraction of bioavailable arsenic in contaminated soils includes desorbable non-occluded forms and some arsenic associated with iron, manganese, and aluminum oxides. Chemical methods may be useful tools to evaluate the effectiveness of remediation technologies and provide information for remediation endpoints.

385 Biological Measures of Bioavailability. McMillen, S.J.*, Chevron Research and Technology Company, Richmond, CA. Results and discussion topics around biological measures of bioavailability from the Pellston Contaminated Soils Workshop will be summarized. Soil organisms may take up chemicals from soil by both passive and active processes. Passive processes include diffusion through cell membranes along chemical gradients, movement with water, or incidental ingestion of contaminated soil particles. This paper will critically review the state-of the art concerning uptake mechanisms for organic compounds and metals by/on plants and soil dwelling organisms as well as the methods currently available for quantifying these mechanisms. The impact of complex mixtures of contaminants and contaminant-nutrient interactions on toxicity will be reviewed. This paper will explore unifying theories such as critical body burden which seek to examine trends across species. If appropriate methods are lacking, recommendations will be provided for how tests could be constructed to provide the required information.

386 Proposed Annex to the ASTM Standard Guide E1841-96, a Standardized Plant Bioassay for Contaminated Soil Materials. J.M. Roper*, ASCI Corporation, Vicksburg, MS; R. A. Price, and J.W. Simmers, USACE Waterways Experiment Station, Vicksburg, MS. A detailed description of the method developed at the Waterways Experiment Station (WES) to determine potential bioavailability and mobility of contaminants from soil to vegetation will be described. The test utilizes *Cyperus esculentus* (yellow nutsedge) as an index plant for a growth period of 45 days. The WES has successfully used this method to predict plant uptake of metals from dredged material placed in upland environments. The method has also been used to evaluate plant uptake of explosives, organotins, and polyaromatic hydrocarbons from various soil materials. This procedure is currently a proposed addition to the ASTM Standard Guide E1841-96: Conducting Renewal Phytotoxicity and Bioaccumulation Tests with Freshwater Emergent Macrophytes. Anticipated publication is in the 1999 ASTM Standards.

387 Composing a Limited Test Battery for Soil Based on the Test Performance and Intrinsic Boundary Conditions of 20 Bioassays. Bierkens, J.*, Klein, G., Corbisier, P., Goyvaerts, M.-P., Van Den Heuvel, R., Verschaeve, L., Weltens, R., Schoeters, G., Flemish Institute for Technological Research - VITO, Mol, Belgium. The test performance (sensitivity, variability,..) and intrinsic boundary conditions (expense, test duration, simplicity,..) of 20 bioassays has been evaluated in response to seven environmental chemicals. Apart from lethality and reproductive failure in earthworms, springtails, nematoda, algae and vascular plants, the endpoints also included bioavailability of metals (bacteria), heat-shock induction (nematodes, algae), DNA damage (bacteria, earthworm, vascular plants), β-galactosidase (*Daphnia*) and esterase activity (algae) and a range of immunological parameters (earthworm). The test substances i.e. cadmium, phenol, pentachlorophenol, malathion, endosulphan, atrazine and trifluraline - each representing a different toxic mode of action - were applied in a dilution series (from 1 mg/kg up to 1000 mg/kg) onto OECD standard soil. The tests were performed both on these artificially contaminated soil samples and on aqueous extracts subsequently obtained from these soils. The aim of the study was to derive a limited but representative, cost-effective and quantitative test battery for soil. A clear distinction in the ranking of the bioassays solely based on their test performance or based on their intrinsic boundary conditions has been calculated. Combination of both set of criteria allowed the selection of a limited and cost-effective number of bioassays with sufficient discriminatory power and reduced uncertainty to assess soil quality. These included lethality and loss of biomass in earthworm, seedling emergence and loss of biomass in cress, growth inhibition in unicellular algae and bacterial biosensors. Although being very sensitive more optimization and normalization is required to include more sophisticated biological indicators such as stress protein induction or immunotoxicity in a cost-effective test battery.

388 Soil Contamination Evaluation and Food-Chain Analysis for Terrestrial Vertebrates: A Stone Age Tool for the Next Millenium. Linder, G.,* HeronWorks, Salem, OR and Oregon State University, Corvallis, OR; Pascoe, G., EA Engineering, Science, and Technology, Bellevue, WA; Fairchild, J., USGS, ECRC, Columbia, MO. With increasing frequency, food-chain analysis is applied to exposure assessments for chemicals in soil. In general, all food-chain models follow this structure:

$$IR_{chemical} = \sum[(C_i \times M_i \times A_i)/BW]$$

where,

$IR_{chemical}$	=	species-specific total rate of intake of chemical by ingestion (mg/kg-day), or dose;
C_i	=	chemical concentration in medium *i* (mg/kg in soil, water, and various dietary constituents);
M_i	=	rate of ingestion of medium *i* (kg/day);
A_i	=	biological transfer factor for chemical in medium *i*; and
BW	=	body weight of receptor species (kg).

Although biological processes such as bioaccumulation are conceptually similar in terrestrial and aquatic habitats, exposure assessments in these habitats vary because of differences in routes of exposure and life history attributes of the receptors of concern. For contaminated soils, simple food-chain analysis generally focuses on dietary exposures to birds and mammals where *partial doses* derived from food, water, and coincidental or intentional soil ingestion contribute the most to a receptor's daily dose. Alternative routes of exposure -- dermal or cutaneous uptake and inhalation -- are often considered of lesser significance except when a chemical's physicochemical properties justify closer analysis of these routes. Here, field and laboratory data from soil contamination studies will be used to estimate daily doses for metals and metalloids across different soil types, then daily dose estimates will be compared to available toxicity reference values. More importantly, critical food-chain model parameters that require much needed research will be identified in order to refine food-chain analysis as a tool for evaluating ecological risks associated with contaminated soils.

389 Soil Ecosystem Function and Assessment Endpoints to Support Generation of Soil Criteria. Wentsel, R.S.*, U.S. EPA, Washington, DC, Kupperman, R.G., U. S. Army, Edgewood, MD, Fairbrother, A., e p & t, Corvallis, OR. Literature on soil ecology and chemical impacts on terrestrial biota is being generated from the microscopic, microbial scale to the macroscopic, ecosystem scale. This material has not been well integrated and synthesized across scales for direct application to the ecological risk assessment process. Nevertheless, there is a growing desire to integrate chemical and/or biological tests of bioavailability and to set risk-based soil criteria for assessment and clean-up goals. This paper will review the tools available for addressing community structure and ecosystem function of both below- and above-ground terrestrial ecosystems. Differences in sensitivity of various parts of the soil ecosystem to different classes of compounds (chlorinated hydrocarbons, polycyclic aromatic hydrocarbons, metals, etc.) will be discussed. All of the material will be consolidated and placed into a risk framework in order to understand the magnitude and shortcomings of our ability to predict the effects of contaminants on the soil and terrestrial ecosystems.

390 Risk Assessment and Risk Management of Contaminated Soils. Menzie*, C.A., Menzie-Cura & Associates, Chelmsford, MA. Development of cost-effective risk assessment clean-up strategies to protect the diversity of ecological receptors and ecosystems is critical. Risk assessments and environmental management of terrestrial systems is complicated by issues of property rights and identification of appropriate receptors and endpoints. Risk assessment and management will be discussed in the context of current and future regulatory drivers. For example, there is a growing desire among the regulatory community to set soil quality guidelines similar to those used for protection of aquatic resources. Similarly, the OECD is pursuing a hazard classification scheme for materials in commerce and transport. All of these issues must be placed in the context of the heterogeneity of soil types (and associated ecosystems) on small and large spatial scales. Furthermore, some terrestrial organisms migrate large distances, integrating exposures across many different ecosystems. This paper will attempt to synthesize the information presented in the previous papers to examine how scientific knowledge about bioavailability, transport, fate, and effects of contaminants in soil can be used to establish a risk framework protective of below- and above-ground ecological receptors.

391 Development of A Novel Injury Quantification Approach For Organisms Exposed to Ah-Active Compounds: A Case Study of the Fox River and Green Bay, Wisconsin. Iannuzzi, T.J.*, Ludwig, D.F., Exponent, Inc., Washington, DC; Moore, M.L., Bigham, G.N., Exponent, Inc., Bellevue, WA; Blankenship, A.L., Kannan. K., Giesy, J.P., Michigan State University, East Lansing, MI. Halogenated organic compounds that act by binding to the aryl hydrocarbon (Ah) receptor in vertebrates are reportedly additive in their toxicological effects on reproduction in organisms such as fish and birds. These endpoints are particularly important for natural resource damage (NRD) injury assessments due to their link to impacts on populations of organisms. Assessing the incremental injuries to organisms from individual Ah *active* chemicals requires the evaluation of the total Ah activity, for all Ah active compounds present as well as the incremental activity from individual chemicals. A reasonable approach to this problem is to determine the total Ah activity using a bioassay, then to measure the concentration of specific chemicals for which the injury assessment is being performed. Using a toxicity equivalence scheme, measured concentrations of individual chemicals can be used to estimate their incremental contribution to the total Ah activity. Alternatively, an attempt to estimate total Ah activity can be made by individually measuring the concentrations of all Ah-active chemicals. There are two key limitations to this latter approach. First, many of the chemicals that exhibit Ah activity have not been identified, and the contributing chemicals vary between sites. Thus, the total analytically determined Ah activity may substantially underestimate the actual activity. Second, the analytical work associated with measuring multiple chemicals is often cost prohibitive. The results of bioassay investigations for fish and birds from the Fox River and Green Bay suggest that the injuries associated with Ah activity are the result of a variety of compounds.

392 **Evaluating a Probabilistic Approach to Hazard Assessment for PCBs and Ecological Receptors for Natural Resource Injury Assessment.** Giesy, J.P.*, Michigan State University, East Lansing, MI; Ludwig, D.F., Iannuzzi, T.J., Exponent, Landover, MD; Moore, M.L., Exponent, Bellevue, WA. Under regulations for natural resource damage assessment, only incremental effects associated with a release are compensable. The requirement for determining incremental damage is technically challenging, particularly for organohalogen compounds that share modes of action and are often found in the environment, but originate from different sources. One technical approach that might contribute to effective injury assessment would be the development and application of distributions of toxicity thresholds among the species of fish and wildlife that provide key natural resource services. Distributions could be employed for injury or uncertainty evaluation. The state-of-the-science and policy foundations for risk and injury assessment are moving rapidly toward full application of probabilistic methods. Most frequently, probabilistic analysis is confined to the exposure side of the risk equation, employing distributions for such parameters as area use, ingestion rate, dietary composition, contaminant concentration, and body weight. It would be useful to ascertain whether scientifically sound probabilistic analysis of the hazard side of the equation is possible in some cases for PCB exposure of wildlife species. In this presentation, we present methods for and the findings resulting from an analysis of the distributions of effects of PCBs to ecological receptors. Receptors are grouped within taxonomic categories and feeding guilds based on the available toxicological data. Distributions of effect doses are developed for each category. Within this framework, variability in toxicity is demonstrated to be a normal consequence of biological differences in susceptibility among taxa. Potential application of some of the resulting distributions for quantitative risk assessment is illustrated.

393 **Injury Endpoint Selection in Natural Resource Damage Assessment.** Lipton, J., Hagler Bailly Services, Boulder, CO. The objective of the injury assessment phase of natural resource damage assessments (NRDAs) is to determine the nature and extent of adverse effects to natural resources that have been caused by releases of hazardous substances. Selection of appropriate injury endpoints is an important element of the injury assessment process. Certain endpoints (e.g., fish or wildlife kills) are readily communicated and accepted. Other endpoints, particularly sublethal endpoints (e.g., effects on growth, reproduction, behavior, histology), are of ecological relevance but can be difficult to describe or quantify in terms that are amenable to damage determination or restoration scaling. This presentation examines the process of endpoint selection, reviews the use and appropriateness of different injury endpoints, and discusses new approaches to quantifying sublethal injuries in a manner that provides a nexus to restoration.

394 **Determination of Injury to Fish Populations: NRDA at CERCLA Sites.** Ripley, B. J.*, and Jenkins, K.D. JSA Environmental, Long Beach, CA. This paper addresses several technical problems that are often associated with the injury determination process at CERCLA sites and discusses strategies for addressing these problems. We deal primarily with the selection and interpretation of measures of injury to biological resources. Studies in the injury determination phase of an NRDA focus almost exclusively on individual organisms while the injury quantification phase is based on population level data. In this paper we will examine examples of measures of injury employed in recent injury determinations studies. Matrix population models will be used to evaluate the relevance of the data from these injury determination studies on individual organisms to the population level measures that are the basis of the injury quantification. The results of these studies will be reviewed and specific recommendations will be made for improving the methods used in the injury determination phase of NRDA at CERCLA sites.

395 **Use of a Bioaccumulation Model for DDE and PCBs in Birds as a Diagnostic Tool for Pathway Determination in NRDAs.** Glaser, D.*, Connolly, J.P., Quantitative Environmental Analysis, New Jersey. Transport pathways of DDE and PCBs to three species of birds were characterized as part of the Southern California Bight Natural Resource Damage Assessment. Extensive field studies were conducted to characterize the dietary composition, foraging behavior, as well as contaminant levels in the eggs of the species of interest and in their prey. This information was used to compute the proportion of the contaminant dose that originates within the Bight: 70 to 80 percent for the peregrine, and 90-95 percent for the bald eagle. Next, bioaccumulation models for DDE and PCBs were developed for the bald eagle and peregrine falcon. Measured contaminant levels in predator eggs were found to be quantitatively consistent with measured levels in its prey, based upon principles of toxicokinetics, both for the peregrine and the bald eagle. This provides support for the estimates of dose that originates within the Bight. An additional model was constructed for the double-crested cormorant. The patterns of movement of the cormorants on Anacapa Island that were based upon natural history information resulted in a relationship between measured contaminant levels in prey and in the cormorant that was consistent with toxicokinetics. In contrast, cormorants from Santa Barbara Island probably feed less intensively in the more contaminated regions of the Bight than previously thought.

396 **Determining Ecological Values as a Basis for Integrating Ecological Risk Assessments and Natural Resource Damage Assessments.** Reagan, D. P., Woodward-Clyde Consultants, Denver, CO. Ecological risk assessments (ERAs) and natural resource damage assessments (NRDAs) are similar in that both processes evaluate exposure pathways and contaminant effects on biota, and both may involve extensive field sampling. Opportunities therefore exist to conduct efficient and cost effective assessments by integrating the two processes. Because ERAs identify Assessment Endpoints (values to be protected) and NRDAs evaluate injury and lost services to all natural resources, integration requires a common basis for evaluation. A systematic identification of ecological values can provide such a basis. The approach begins by identifying healthy ecosystems as the ultimate ecological value, then proceeds to identify lower levels of ecological organization, based on the interrelationships among functional components of the ecosystem(s) being evaluated. Physical resources (air, water, sediment, soil, etc.) are added in the context of the services they provide to biotic components of their respective ecosystems. This approach provides a common basis for determining assessment endpoints and for identifying potentially injured resources and ecological service losses. It therefore provides a means of addressing natural resource damage assessment issues cost-effectively by maximizing the use of existing information and by reducing or eliminating the need for separate injury/damage assessment investigations. Examples for various ecosystems are presented.

397 **Using the CERCLA/RCRA Required Ecological Risk Assessment to Avoid Separate Natural Resource Damage Action.** Maughan, J, CH2M HILL, Boston MA. There is concern among many potentially responsible parties (PRP) that after a removal or remediation plan has been selected and approved, subsequent Natural Resource Damage (NRDA) claims could force a *Clean Up 2*. This is true because in some cases the remediation plan can achieve risk reduction that is

protective of human health and the environment, and thus meet the requirements of CERCLA or RCRA, and not restore the natural resources, as required by NRDA. There could also be historic lost natural resource services that are not addressed by the remediation. In some cases the remediation could even cause natural resource damage, thus adding to the PRP's natural resource liability. Although the CERCLA or RCRA ecological risk assessment is different from a NRDA, often, slight modifications to the assessment can eliminate the need for a separate NRDA action. Modifications might include incorporating trustee's NRDA concerns as assessment endpoints so that injury and lost service to these resources are considered in the risk assessment. Minor modifications can also be made to predict risk (and thus injury and lost service) to the endpoints (and thus natural resources of concern) after implementation of the alternative remediation option. In this manner the residual damage and applicability of NRDA, can be judged prior to final selection of the remediation option. The process can be performed in an iterative fashion to modify alternatives so that there is no need for separate NRDA action. In cases where this approach is applicable the lengthy, expensive, and painful process of a formal NRDA is avoided. The need for a second clean up can also be avoided and in some cases there is no need for a Natural Resource Damage settlement.

398 **The Noaa Coastal Resource Coordination Program: Cooperatively Resolving Natural Resource Liability Through the Remedial Process.** Fritz, A.T.*, NOAA, Seattle, WA; Matta, M.B., NOAA, Seattle, WA; Lindsay, J., NOAA, Seattle, WA. Resolving liability for natural resource injury at hazardous waste sites can most efficiently be accomplished through the cooperative efforts of natural resource trustees, cleanup agencies, and responsible parties. CERCLA authorizes natural resource trustees to grant covenants-not-to-sue for natural resource liability if appropriate measures are taken to protect and restore natural resources. Assessing injury as early as possible by conducting effective ecological risk assessments can allow liability for natural resource injury to be resolved as part of a global settlement for all CERCLA liability. Effective ecological risk assessments support the selection of remedies that will protect natural resources and provide information useful to scale restoration of injured resources. If such measures to protect and restore natural resources are agreed to as part of global settlement negotiations, responsible parties can be efficiently released from liability. Although incorporating injury assessment into ecological risk assessments may be thought of as an "advance", NOAA's Coastal Resource Coordination (CRC) Program has been working to cooperatively achieve protection and restoration of injured coastal natural resources since 1984. Agreements to restore natural resources have been achieved at more than 25 sites, and protection for coastal natural resources has been improved at more than 300 sites. The approach includes working with lead cleanup agencies (primarily EPA), other natural resource trustee agencies and tribes, and responsible parties to ensure that adverse effects to natural resources are evaluated in ecological risk assessments. Where potential injuries of interest to trustees cannot be incorporated into ecological risk assessments, separate investigations may occur. As the benefits of reaching cooperative settlements that protect and restore natural resources become apparent, there is increasing pressure on EPA and trustee agencies to collect information to allow them to achieve a coordinated cleanup and restoration.

399 **Prospective Ecological Risk Assessment for Risk/Benefit and Risk Management Evaluation of Proposed Wetlands Restoration.** DeMott, R.P.*, Jones, H.D., Schell, J.D., ATRA Occupational & Environmental Services, Tallahassee, FL. Lake Apopka in Central Florida has been impacted by intensive commercial muck farm agriculture along its shores. As part of the overall strategy for rehabilitating the lake, restoration of historical wetlands in such areas has been proposed. Expected environmental benefits include reductions in nutrient and agrochemical input and expansion of the wetlands ecosystem filtering the water entering the lake. However, federal authorities requested a quantitative ecological risk assessment to evaluate the potential local risks associated with flooding the agricultural lands and allowing a wetlands community to develop. Based on sampling and screening analyses, organochlorine compounds formerly used in the fields were identified as the primary risk factors. The goal was to determine the extent of potential future risks after flooding and characterize the effectiveness of various risk management strategies. A prospective risk model was developed from receptor use patterns in nearby wetlands. Benthic, aquatic, and piscivorous receptors were evaluated and relevant toxicity-related doses and/or concentrations were developed. Locally derived biota-sediment accumulation factors were used to estimate bioaccumulation. Risks were characterized with standard ecological quotients (EQs). The EQs for several receptor/chemical combinations exceeded 1 and were evaluated in a weight-of-evidence analysis considering available risk management strategies and factors expected to reduce concentrations over time. Even the highest modeled EQ (13 for great blue heron/DDT), was expected to be rapidly reduced by sedimentation and anaerobic degradation. Further, the ability to isolate and monitor the restored area can ensure that the conservative exposure factors of the prospective model are not met. The quantitative nature of both the assessment and demonstration of the suitability of management plans facilitated regulatory approval for this restoration plan.

400 **Controlling Nrda Through Pre-incident Planning.** Robertson, S.B., Woodward-Clyde International, Los Angeles, CA. The Oil Pollution Act of 1990 (OPA 90) made it clear that the party(ies) responsible for an oil spill should compensate the public for injuries to natural resources. Trustees of these resources are to assess damages and ensure that restoration takes place to make the public whole again. Recommended procedures for Natural Resource Damage Assessment (NRDA) have been promulgated by NOAA (National Oceanic and Atmospheric Administration, Final Rule, January 5, 1996). This rule requires the trustees to invite the responsible party to participate in the process. Through pre-incident preparations and planning of actions to be taken in the event of a spill, the responsible party can be better prepared to be an active player in this cooperative effort. The cost of damages may not go away, but at least they may be limited to reasonable and realistic levels. Examples of pre-incident activities include reduction of impact by preplanning appropriate spill response actions, awareness or collection of baseline data, establishment of relationships with trustees and consultants, setting up an NRDA team and its relationship with the Incident Command System (ICS) response team, preparations for the prompt collection of ephemeral or time-critical data, gathering appropriate study protocols to ensure scientifically valid data, and including NRDA as a regular part of spill drills.

401 **From Mud to Management: Using the Sediment Quality Triad Approach in Tampa Bay, FL.** Greening, H.S.*, Tampa Bay Estuary Program, St. Petersburg, FL; Long, E.R., NOAA, Seattle, WA; MacDonald, D.D., MacDonald Environmental Services, Ltd., Ladysmith, British Columbia, CAN. Results from sediment chemistry and toxicity tests conducted by NOAA and others in the 1980s and early 1990s in Tampa Bay indicated that, although much of the bay is relatively uncontaminated, several *hot spots* of sediment contamination exist, associated with urban/industrial runoff and/or large marinas or ports. Contaminants of concern (COCs) have been identified using a combination of sediment quality guidelines and risk assessment methods, and consist primarily of metals, PAHs, PCBs and pesticides. Local, state and federal partners working through the Tampa Bay Estuary Program have adopted the use of the sediment quality triad approach (sediment chemistry, toxicity, and benthic invertebrate community structure) as the primary tool to provide a means of estimating the relative risks to the benthic community and to human health of contamination levels in *hot spot* areas. Specific numeric sediment concentration targets (identified as *clean)* have been recommended for some COCs (i.e., concentration <sediment quality guidelines developed for Florida estuaries), and *clean* targets for measures of toxicity (i.e., not significantly toxic to amphipods or sea urchin fertilization) and benthic community structure (i.e., indicator species [crustaceans] present) have been identified by the TBEP partners. Several questions remain, including whether each leg of the triad should receive equal weight during interpretation. Results of the analyses are

being used to prioritize management actions in the watersheds draining to highly contaminated areas, focusing on sources of those COCs in the sediments which do not currently meet agreed-upon targets.

402 Relationship Between *Ampelisca* Sediment Toxicity Test Response and Parameters of Benthic Community Condition. Scott, K.J., Science Applications International Corporation, Narragansett, RI. Amphipod 10-day solid-phase toxicity tests are commonly used to assess the extent of sediment contamination and chemical bioavailability. Tests with the tube-dwelling amphipod *Ampelisca abdita* are employed in the CoE dredged material evaluation program and in monitoring programs to assess bioeffects and estuarine condition by NOAA and EPA EMAP and the mid-Atlantic Integrated Assessment (MAIA), among others. Survival of *Ampelisca* in this test was compared to various parameters of benthic community condition that included but was not limited to the EMAP Benthic Index, abundance of amphipods and other crustaceans, and species diversity. Specific programs that were investigated were EMAP, MAIA, Navy New England risk assessments, and New Bedford Harbor evaluations. Survival in toxicity tests was most predictive of community response in more highly contaminated sediments in EMAP-Virginian Province, Navy, and superfund sites. The level of confidence in this acute response decreased as did the level of contamination in EMAP-Louisianian and Carolinian Province sites. These analyses support the contention that test responses are predictive of responses *in situ*, particularly when compared to the presence and abundance of pollution sensitive species. They also indicate the level of test toxic response (i.e., survival) which is most predictive.

403 Relationships Between Benthos and Sediment Toxicity in San Francisco Bay. Thompson, B.*, San Francisco Estuary Institute, Richmond, CA; Hunt, J., and Anderson, B., University of California, Santa Cruz; Oakden, J., Moss Landing Marine Laboratory, Moss Landing, CA; Taberski, K., San Francisco Bay Regional Water Quality Control Board, Oakland, CA. *Reference* benthic assemblages have been identified in San Francisco Bay and are generally associated with low sediment contamination (using mean ERM quotient), but those sediments occasionally indicate toxicity to the amphipod *Eohaustorius estuarius* and / or larval bivalves. Benthos may be moderately to severely impacted by contamination in localized areas of the Bay. Benthos near major municipal outfalls appear only slightly impacted, as evidenced by elevated numbers of species, but are not always toxic. Other sites with high contamination that are toxic have only moderately impacted benthos. A few sites with severe contamination have no benthos, and are always toxic. Amphipod toxicity generally corresponds to reduced numbers of amphipods in the benthos, but the relationship is confounded by differing life histories of the several species of benthic amphipods collected in samples. For example, abundances of *Ampelisca abdita* are higher in the summer than in the winter. Amphipod abundances are generally lower at sites with more sand. Amphipod survival in laboratory tests is seasonal at some sites, being higher in the wet periods when more contaminants are flushed into the Estuary than in the dry season. All of those factors must be considered when attempting to relate benthic parameters to sediment toxicity. No consistent relationships were observed between benthic parameters and results of the larval bivalve elutriate test.

404 Chronic Toxicity and Benthic Effects Along Southern California Municipal Wastewater Exposure Gradients. Bay, S.M.*, Greenstein, D.J., Jirik, A.W., and Brown, J.S., Southern California Coastal Water Research Project, Westminster, CA. Sediments near large ocean discharges of municipal wastewater in Southern California are not acutely toxic, even though they are enriched in many contaminants and contain altered benthic communities. Long-term sediment exposures (4-16 weeks) were conducted to investigate the relationship between growth impairment in three species (brittlestar, sea urchin, and amphipod) and benthic responses at selected discharge sites. The test methods included a surrogate species (amphipod) and a local species (brittlestar) having reduced population size near the discharges. There was generally a poor predictive relationship between the laboratory and field data. Benthic responses (e.g., reduced brittlestar abundance and altered community composition) were more sensitive to the contamination gradients than were the toxicity tests. Predictive ability of the toxicity tests was not improved by increasing the length of exposure or using a species (brittlestar) known to have an adverse population response at the study sites. The best correspondence between lab and field effects was obtained for highly contaminated sediments from the Palos Verdes Shelf. Sediment contaminant concentrations were relatively low at some sites that lacked a toxicity-benthos relationship, indicating that benthic effects were due to factors that were not accurately reproduced in the laboratory.

405 Coupling Sediment Contamination and Toxicity Testing to Benthic Community Responses off a Marine Outfall in Western Portugal: Do We See Only What We Want To? Quintino, V.* and Rodrigues, A. M., Universidade de Aveiro, 3810 AVEIRO, PORTUGAL. In order to evaluate the responses of the receiving system and to suggest the degree of treatment needed for an outfall facility off the western coast of Portugal, pre- versus post-operation environmental studies were undertaken. The programme, as a whole, included chemical and toxicological characterization of the effluent, the receiving water and sediments, biology studies of the plankton, the fish and the benthic communities, microbiological analysis and a 3-D model of the plume dispersion. In this presentation, we focus on the sediment component, for which a sediment quality triad approach was used, including measures of physical and chemical descriptors, acute and chronic sediment toxicity tests, using a sea-urchin and an amphipod species, and the composition and structure of the resident macrofauna. This information was collected from 20 sites, in March 1994 and April 1997. Discharge was initiated May-June 1994. During 1997, the benthic communities and occasionally sediment toxicity, were also studied every 3 months in some of the previous sites, in order to evaluate the temporal variability of this system. The results obtained so far allow the identification of both the biotic and abiotic gradients in the area, their close relationships and the connection to the broarder seascape where the study is being undertaken. In terms of responses to the discharge, the results indicate the milder reaction of a coastal system dominated by fine clean sand to an organic enrichment process, from which persistent contaminants are basically absent. Although the elutriate based bioassays show a more difficult pattern to fit in with this image, acute and full-life cycle amphipod tests reveal a close relation with the other descriptors. It will be argued that, relative to this type of disturbance and situation, only from full-life cycle tests can we gain insight into the relationship with benthic communities responses.

406 Use of the Sediment Quality Triad Approach to Evaluate Benthic Invertebrate Effects with Toxicity Tests and Sediment Chemistry. Canfield, T.J.*, R.S. Kerr Environmental Research Laboratory, U.S. Environmental Protection Agency, Ada, OK., Dwyer, F.J., Ingersoll, C.G. Kemble, N.E., Environmental and Contaminants Research Center, U.S. Geological Survey, Columbia, MO. Sediments from three Great Lakes Areas of Concern (AOC), the Clark Fork River superfund site (Montana), the upper Mississippi River, and Turkey Creek (Joplin, Missouri) were evaluated for levels of contamination using sediment chemistry, laboratory toxicity tests, and benthic invertebrate community structure assessments. Good concurrence was evident between measures of laboratory toxicity, contaminant concentration, and benthic invertebrate community composition in extremely contaminated samples. In moderately contaminated samples, less concordance was observed between the benthos community composition and either laboratory toxicity test results or sediment chemistry. However, there was good concordance between laboratory toxicity results and sediment chemistry in moderately contaminated samples. Laboratory sediment toxicity tests may better identify chemical contamination in sediments than many commonly used measures of benthic invertebrate community composition. Spatial and temporal variation of contaminants in sediment may additionally influence distributions of benthic communities. Benthic measures may also reflect other factors such as habitat alteration which affect community composition in addition to or in spite of chemical contamination as expressed by laboratory toxicity test results. The dynamic nature of benthic invertebrate community composition may be influenced by abiotic factors such as grain size, total organic carbon, dissolved oxygen, and biotic factors such as inter-species and intra-species competition as well as predator prey interactions regardless of sediment contaminants. Because of these other factors, contaminant effects may only be observable in the benthic invertebrate community in the most contaminated sites. Controlled field studies using colonization studies of contaminated sediments are needed to better evaluate benthic community responses to contaminants associated with sediments.

407 Ecological Effects of Sediment-associated Contaminants in Inner Burlington Harbor, Lake Champlain. Diamond, J.*, Richardson, A.L., and Daley, C. Tetra Tech, Inc., Owings Mills, MD. This project: (1) analyzed and compared current sediment and benthic ecological conditions in the harbor with data collected previously; (2) determined the potential for chronic sediment quality effects on biota in the harbor; and (3) evaluated potential long-term risks to aquatic biota in the harbor relative to developing management alternatives. Twenty sites (10 reference [relatively clean] and 10 possibly impaired) were sampled in summer 1997 for whole sediment toxicity, polynuclear aromatic hydrocarbons (PAHs), select metals, several physicochemical parameters, vertical profile characteristics, organism tissue PAHs, lead, protein expression (biomarkers), and benthic macroinvertebrate community integrity. Fathead minnow growth in sediment toxicity tests corresponded reasonably well with benthic macroinvertebrate data and both the fish and *Hyalella* 10 day tests correctly predicted areas of highest contaminant concentrations. *Hyalella* was more sensitive to sediment characteristics than the fish and generally predicted greater impacts than measured using benthic macroinvertebrate analyses. Chronic larval fish survival and growth in laboratory tests were related to SEM/AVS while *Hyalella* survival was related to zinc and lead concentrations. Benthic assemblage integrity was related to sediment PAHs and to a lesser extent, metals such as copper and nickel. The spatial pattern of contaminants in the harbor was consistent with earlier results, however, there were two striking changes: (a) significantly lower concentrations of most contaminants in surficial sediments presently as compared to 3-4 years ago and (b) a substantial increase in the number of zebra mussels in the harbor. The decrease in sediment contaminants is coincident with the relocation of a sewage outfall. Zebra mussels were associated with macrophytes and near shore areas. Interpretations of benthic biological data were limited because of highly heterogeneous sediment conditions in the harbor and the lack of clear reference sites. Follow-up chronic toxicity, bioaccumulation, and protein expression testing at select sites sampled in the spring 1998 will be used to discern long-term risk potential and causes of risk to aquatic fauna in the harbor.

408 Application of a Micro-Scale Bioassay Set for the Evaluation of Sediment Quality. Ahlf, W.*, Fretwurst, S., Gratzer, H., TUHH, Hamburg, Germany, Traunspurger, W., University of Bielefeld, Bielefeld, Germany. A three-year study was performed to validate recently developed sediment toxicity tests. The methods used are rapid and inexpensive. Among the tests are four (Microtox™ and algae with elutriates in microplates, bacteria and nematodes as solid-phase tests), which provide a basic set of bioassays for the assement of the rivers Elbe and Rhine. The objective of the study was to quantify toxic effects for a sediment quality ranking. About 250 sediment samples were collected and evaluated. More than 90 % of the samples caused toxic effects in the bioassays. Geochemical characterizations, microbial activity, nematode size classes, and macroinvertebrate community assessments were conducted on parts of the collected samples. We have observed site-specific responses which are related to TBT-contents and particle size distribution. The paper will focus on the interpretation of bioassay results in comparison of variation in space, time and sediment composition. The relationship between toxicity tests and sediment chemistry /biological field data will be discussed in detail.

409 Evaluation of the Effect of Land-use Activities on Sediment Quality Within the Galveston Bay System. Guillen, G.J.* TNRCC, Houston, TX; Carles, L., TNRCC, Houston, TX; Broach, L., TNRCC, Houston, TX; Smith, S. A sediment quality assessment of Galveston Bay was conducted during 1997. The objective of this study was to determine the relationship between various land-use activities and sediment contaminant levels, toxicity and benthic communities. A total of fifty stations representing a gradient of minimally impacted to severely impacted sites were selected. Various sites were selected adjacent to various shoreline land-use activities. These activities including shipyards, refineries, chemical plants, oil-field facilities and marinas. Several sites included active wastewater discharges. A sediment triad approach which utilizes concurrent bioassay, chemical and community measurements of sediment samples was selected to assess the relative degree of contamination at each site. Several bioassays including the Microtox™ solid-phase test, and, two elutriate tests including the mysid shrimp *Mysidopsis bahia*, chronic and acute test, and sheepshead *Cyprinodon variegatus* acute test were conducted on samples collected at each site. Concurrent chemical measurements were conducted on sediment samples including inorganic (metals) and organic priority pollutant compounds, SEM, AVS, TOC and sediment particle size distribution. Benthic community samples were also collected at the same locations. The modified EPA EMAP benthic index for estuaries was generated for each benthic community sample. Based on the results of this survey trends in sediment contamination based on chemical, bioassay and community analyses appeared to be strongly related to land-use activity. In addition, broader patterns in contamination associated with watershed scale land-use activities were also observed. Highly industrialized watersheds contained the largest number of contaminated sites.

410 Relationship Between Sediment Toxicity and Benthic Community Degradation in California Coastal Waters. B. Anderson*, J. Hunt, B. Phillips, R. Tjeerdema, University of California, Santa Cruz; R. Fairey, J. Oakden, San Jose State University; M. Puckett, California Department of Fish and Game; E. Long, National Oceanic and Atmospheric Administration (NOAA). Sediment contamination and bioeffects were assessed in southern California bays, harbors, and estuaries by the State Water Resources Control Board Bay Protection and Toxic Cleanup Program and NOAA. Monitoring relied on the Sediment Quality Triad approach incorporating measures of chemistry, toxicity, and benthic community structure. Toxicity tests included amphipod survival, molluscan and echinoid development, and polychaete growth and survival. Benthic community structure was assessed using a Relative Benthic Index, calculated based on measures of the total number of fauna, number of crustacean species, and numbers of positive and negative indicator species. Univariate and multivariate statistical analyses indicated significant positive relationships between increased survival of amphipods in laboratory toxicity tests, greater numbers of crustacean species, and a higher Relative Benthic Index. There was a positive relationship between increasing percentages of normal development of mollusc larvae in laboratory toxicity tests and greater numbers of mollusc species and individualsin the benthos. There were significant negative relationships between increased chemical contamination and measures of both toxicity data and benthic community diversity and abundance. Toxicity data used in conjunction with chemical measures, physical measures, and information on other environmental factors which influence benthic community structure were found to be useful in identifying sites where chemical contamination may have resulted in ecological impacts. These data will also be used to illustrate specific examples of situations where, for a variety of reasons, toxicity tests and benthic community analyses need to be interpreted cautiously.

411 A Fingerprinting Misinterpretation: Previous Claims for the Natural "Petroleum" Hydrocarbon Background in Subtidal Sediments of Prince William Sound, Alaska. Short, J. W.*, Wright, B. A., Larsen, M. L., Holland, L. G., National Marine Fisheries Service, Juneau, AK; Kvenvolden, K. A., Carlson, P. R., Hostettler, F. D., Rosenbauer, R. J., U. S. Geological Survey, Menlo Park, CA. Claims have been made repeatedly in the literature that subtidal sediments in Prince William Sound (PWS), Alaska were contaminated by crude oil produced from natural seeps prior to the 1989 *Exxon Valdez* oil spill. This hydrocarbon background was identified with natural oil seeps at Katalla and elsewhere along coastal areas of the Gulf of Alaska (GOA), based on the presence of oleanane and the ratio of C_2-dibenzothiophenes to C_2-phenanthrenes. A more thorough assessment of all available hydrocarbon data combined with an evaluation of other plausible hydrocarbon sources reveals the actual source is most likely coal. This evidence includes (1) high concentrations of total PAH in continental shelf sediments co-extensive with upland coal deposits; (2) low ratios (<0.2) of triaromatic steranes to methylchrysenes in sediments and coals, but high ratios (11 and 13) in Katalla oil; (3) the presence of bisnorhopane in sediments and coal, but its absence in Katalla oil; (4) the bioaccumulation of PAH in biota collected within 100 m of the Katalla oil seeps, but the absence of most PAH in mussels collected near coalfield drainages 9 km from the oil seeps; (5) weathered PAH distributions that characterize oil from Katalla seeps but are not evident in benthic sediments, and (6) absence of PAH losses from experimentally weathered coal. PAH in these coals are not bioavailable, so the presence of coal in the benthic sediments of this region confers no adaptive benefit to biota of the aquatic ecosystem with respect to PAH insults from anthropogenic sources.

412 Spilled Oil Persistence in the Marine Environment: Petroleum Chemistry and Evaporation/Dissolution Weathering. Payne, J.R., Payne Environmental Consultants, Inc., Encinitas, CA. To evaluate changes in the physical and chemical properties of crude oil spilled in subarctic open-ocean conditions, replicate outdoor flow-through seawater wave-tank experiments were initiated under summer and winter conditions near Homer, Alaska. Turbulence was continuously introduced into 2,800 L wave tanks by paddle wheels, and the experiments were run for up to 13 months under ambient weather conditions. To simulate wind-driven advection of floating oil over previously unexposed seawater, the flow-through system was adjusted to provide one tank-volume turnover every four hours. Thus, a dynamic (rather than static) equilibrium was maintained. This allowed dissolved/dispersed oil to be removed from the water column by advective processes while ambient levels of nutrients, bacteria, and suspended particulate material were continuously introduced. Physically dispersed oil-droplet concentrations maximized at 9,420 $\mu g/L$ within 5 minutes of the spill and then remained fairly constant at 2500-2800 $\mu g/L$ for 1-4 hours. Between 12-24 hours, oil droplet concentrations were 1300-1700 $\mu g/L$, and they subsequently dropped to 570 ug/L after 2 days. Continued droplet dispersion was limited by increases in oil viscosity, and concentrations declined from 300 $\mu g/L$ after 3 days to 90 $\mu g/L$ after 12 days. Dissolved-phase alkyl-benzene concentrations maximized at 260 $\mu g/L$ after 1-2 hours and dropped to 70 $\mu g/L$ after 12 hours. After 6 days alkyl-benzene concentrations were 7.2 $\mu g/L$, and by day 12 they were < 0.8 $\mu g/L$. Dissolved-phase PAH concentrations maximized at 160 $\mu g/L$ after 4 hours; however, their persistence in the water column was considerably longer, remaining at 25 $\mu g/L$ for 6-12 days. In fact, as long as there was source oil (albeit weathered) on the water surface, total alkylated naphthalenes, dibenzothiophenes, and phenanthrenes persisted at concentrations of 5-7 $\mu g/L$ for 4-9 months.

413 Supercritical Fluid Extraction of Diesel from Weathered Soils. Lee, C. M.*, EE&S, Clemson University, Clemson, SC 29634-0919; Rutten, D. E., U. S. Army, Aberdeen, MD; Danker, R. M., EE&S, Clemson University, Clemson, SC 29634-0919. Soil was collected from four U.S. Army posts, Ft. Carson, CO, Ft. Jackson, SC, Ft. McClellan, AL, and Ft. Sill, OK, and spiked with 100 mg diesel fuel per gram soil to simulate a spill. The spiked soils were divided into two portions with one portion reserved in a freezer as a control and the other weathered on the roof of the EE&S laboratory. The weathered soils and control soils were sampled on a regular basis for six months and after 22 months they were again sampled for six months. Samples were extracted with supercritical carbon dioxide and analyzed by gas chromatography with a flame ionization detector. About 20% of the fuel volatilized during the spiking of the soil samples. The largest percentage of diesel was recovered from the soil with the largest percentage of organic matter (Ft. McClellan, AL, 6.3 % organic matter). Clay to a limited extent influenced the extraction of diesel from the other three soils which contained about the same amount of organic matter (0.9%). The percentage of diesel extracted from the Ft. Carson, CO, soil, which had the largest percentage of clay (37%), declined as moisture content increased due to rain events. Supercritical fluid extraction of the control soils resulted in essentially the same recovery for the study period. Gas chromatography/mass spectromery analysis of the weathered and control soils indicates that the weathering time can be assessed by comparisons of the loss of n-alkanes to tricyclic terpanes.

414 Identification of Non-point Source Related PAHs Through Pattern Recognition. Douglas, G.S.*, Brown, J.S., Camp, W.H., Naughton, G.P. Arthur D. Little, Inc., Cambridge, MA. The presence of polynuclear aromatic hydrocarbons (PAHs) in sediments or soils at urban sites may be atmospherically derived. The presence of PAHs may be indicative of petroleum sources or pyrogenic sources, these PAHs may not be derived from a single source, but from the sum of many anthropogenic sources which contribute PAHs to urban dusts, soils, and sediments. Because the PAHs in urban dusts are primarily pyrogenic in origin it is difficult to differentiate between background pyrogenic PAHs and other sources of pyrogenic PAHs. The distribution of PAHs were examined in urban dust, storm water, and sediments. Analytical methods included quantitation of parent and alkylated PAHs. Several sediment and dust samples, known to contain PAHs from a point source, in addition to background, were also analyzed. The use of Principal Component Analysis (PCA) provided a statistical means to distinguish samples containing "background" pyrogenic PAHs from those effected by a point source and to identify the PAH contribution in urban dust and sediments derived from background sources. This background represents the commingling of many point and non-point source emissions, including industry, incineration, automobile exhaust, and power generation. The risk from a heavily loaded background sample was compared to a petroleum impacted sample from a low background area using a methodology consistent with the TPH Criteria Working Group.

It may be desirable to not define background as a concentration, urban environments create situations where background input can be expected to accumulate and concentrate. Second, comparison of PAH concentrations in site specific samples to a background level as a pattern, and not as individual compounds, may yield results with more meaning. In this study, several samples clearly impacted by a No. 2 Fuel Oil Spill had lower concentrations of PAHs than nearby background samples.

415 Accumulation of PAHs and Hopane/Sterane Biomarkers in Fish, Mussels, and SPMDs as a Means of Discriminating and Apportioning Petroleum Sources. Luellen, D.R.*, Shea, D., N.C. State University, Raleigh, NC. Source discrimination and apportionment of petroleum hydrocarbons in the environment is critical to the prioritization of resource management options and to the identification and liability of potentially responsible parties in environmental restoration and litigation. In surface waters, the primary matrices available for measuring petroleum components are water and aquatic biota. However, the concentrations of petroleum components in water usually are very low and highly variable, and petroleum residues in biota can be modified by selective uptake and metabolism. We hypothesized that semi-permeable membrane devices (SPMDs) might accumulate a distribution of PAHs and sterane/hopane biomarkers that is much more conserved than that found in fish or even mussels. We conducted laboratory studies of the uptake kinetics of over 50 PAHs and over 50 hopane and sterane petroleum biomarkers in fish, mussels, and SPMDs. We also measured these same compounds in fish and SPMDs at several National Wildlife Refuges (NWRs) with active oil production along the lower Mississippi River. Diagnostic ratios calculated from the relative distribution of the PAH and biomarker compounds in the samples were compared to ratios determined from the analysis of the crude oil from the same refuges and to published ratios for diesel and other petroleum products. The ratios do not indicate a single source of contamination on any of the refuges, but on Delta NWR the biomarker ratios indicate the primary source of hydrocarbon contamination is from the on-site oil production. The relative merits and limitations of the alternative approaches will be summarized.

416 **Time-integrated, Flux-based Monitoring Using Semipermeable Membrane Devices to Estimate the Contribution of Industrial Facilities to Regional PCB Budgets.** McCarthy, J.F.*, Southworth, G.R., Palmer, J.A., Oak Ridge National Laboratory, Oak Ridge, TN, Ham, K.D., Washington Department of Fish and Wildlife, Ellensburg, WA. Passive monitoring of polychlorinated biphenyls (PCBs) with semipermeable membrane devices (SPMDs) was used to identify and quantify the contributions of point- and non-point sources to the total PCB flux in a drainage system encompassing three Department of Energy (DOE) industrial and research facilities in Tennessee (USA). PCB concentrations were greatest at discharge points containing process wastes from the DOE industrial facilities. The significance of these releases was evaluated by calculating the PCB flux (mass per unit time) at different monitoring locations, using time-integrated estimates of the aqueous concentrations of PCB, estimates of the volumetric flow rates of discharges, volumetric flows in receiving streams during the deployment period. The total flux of PCBs from the three DOE facilities accounted for most of the total PCBs entering the Clinch River from the streams near the facilities. Principal components analysis was helpful in attributing sources of PCB. In one stream receiving multiple inputs of PCB, congener profiles from upstream sources and effluent discharge were consistent with a mixture of the congener profiles in the downstream receiving water. In another stream with a single upstream source of PCB, changes in PCB flux and congener profiles suggested an apparent steady-state distribution between dissolved PCB and PCB adsorbed to organic matter in the stream bed. Time-integrated, flux-based monitoring can be useful across a range of spatial scales for evaluating the significance of point- and non-point contaminant sources, and can provide information needed to identify and prioritize feasible remedial alternatives.

417 **Development of *In situ* Partition Coefficients for PCB Congeners in the Hudson River.** Garvey, E. A.*, TAMS Consultants, Inc., Bloomfield, NJ; Butcher, J. B., Tetra Tech, Research Triangle Park, NC; Tomchuk, D., US Environmental Protection Agency, Region II, New York, NY. Hudson River PCB contamination dates back to the early 1950s with the onset of PCB-contaminated discharges from two General Electric facilities located near Ft. Edward, NY. As part of the ongoing USEPA reassessment of PCB contamination in the Hudson, water column samples were collected from fifteen stations located in the fresh water portion of the Hudson River and its tributaries. Samples, consisting of 17 liters of water, were field filtered within 4 hours of collection via 0.7 μm glass fiber filter with each portion subsequently analyzed for PCB congeners. The large sample size served to minimize the number of results reported as non-detect. The results were compiled with separate measurements for total suspended solids to yield apparent two-phase partition coefficients for each congener for each sample. Individual sample results were then normalized for particulate organic carbon content to yield a K_{oc}. A second, field-temperature-based correction was also developed from a secondary data source and applied to the data. The results show a good correlation between organic carbon partitioning and temperature, yielding a set of coefficients for 60 individual congeners which can applied over a wide range of river conditions. When particulate-phase concentrations are predicted from dissolved concentrations, the average percent difference between observed and predicted particulate concentrations is $\pm$ 43 percent. Resulting *in situ* partition coefficients show systematic differences from partition coefficients predicted from octanol-water partitioning. *in situ* partition coefficients for low molecular weight congeners were elevated relative to those predicted from octanol-water partitioning. Similarly, *in situ* partition coefficients for high molecular weight congeners were depressed relative to those predicted from octanol-water partitioning.

418 **Toxaphene Quantitation: Improvements and Application to Environmental Samples.** Glassmeyer, S. T, Shanks, K. E., and Hites, Ronald A., Indiana University, School of Public and Environmental Affairs and Department of Chemistry, Bloomington, IN 47405. Toxaphene is a complex mixture of at least 600 hexa- to decachlorinated norbornanes and norbornenes. Like PCBs and other environmentally persistent organochlorines, toxaphene is ubiquitous in the environment because of its atmospheric transport away from areas of use. Toxaphene's complex nature makes accurate quantitation difficult. Quantitation methods using mass spectrometry can be categorized into three types: (a) those which include only a handful of peaks in each sample; (b) those that sum all of the peaks in a particular mass to charge range; and (c) those which compare the ratio of the quantitation to the confirmation ion to determine if a peak is indeed toxaphene. The first two methods under- and overestimate the amount of toxaphene in a sample, respectively, and do not give an accurate homologue distribution, while the third method is time consuming and subject to human error. As an alternative, we have developed a QBASIC program that performs the isotopic ion ratio comparison, thus decreasing the time required for analysis while maintaining accurate and precise quantitation. To test the program, we have examined the atmospheric concentrations near Lake Superior. Air samples were collected from September 1996 to December 1997 at Eagle Harbor, Michigan, a remote shore-side site operated by the Integrated Atmospheric Deposition Network (IADN). Vapor phase concentrations range from over 100 pg/m^3 in the summer months to less than 1 pg/m^3 in the winter. The homologue composition of these samples tended to skew towards the lower chlorinated groups, as compared to a reference standard.

419 **Prediction of Hexachlorobenzene Bioconcentration in Whole Tissues Using Common Headspace Partitioning.** Ferguson, P.L.* and Brownawell, B.J., Marine Sciences Research Center, SUNY, Stony Brook, NY. The potential for hydrophobic organic compounds (HOCs) to bioconcentrate within tissues of aquatic organisms has often been related to the organism's total percent lipid content. The lipid-normalization approach is based on assumptions of bulk-phase partitioning behavior and neglects the potential contributions of other biomolecules such as proteins. It is clear that a substantial portion of the lipid tissue in organisms is arranged in bilayer membranes, which may not approximate a bulk phase with respect to pollutant partitioning. In addition, protein, which may constitute up to 80% of an organism's dry weight, may play an important role in biopartitioning of nonpolar organics. This may be especially true in the case of organisms having low percent lipid tissues. In order to study the dependence of hexachlorobenzene (HCB) biopartitioning on biochemical tissue composition, we utilized a novel common headspace partitioning device. The design of this device allowed for rapid equilibration between water and powdered tissue suspensions through a shared headspace. HCB partitioning into eleven marine organism tissues was studied. Tissue lipid contents varied between 0.7 (marine seaweed *Chondrus*) and 21% (eel liver). The ratio of protein to lipid content in the tissues varied from 2.1 (eel liver) to 36 (sole fillet). Polar phospholipids comprised between 6.7 (marine seaweed *Ulva*) and 83% (*Chondrus*) of the extracted lipid pools. Deviation of HCB lipid normalized tissue/water partition coefficients from published HCB triolein/water partition coefficients may indicate significant contribution of hydrophobic proteins to HOC bioconcentration. These deviations may also indicate effects of variations in the ratio of storage to membrane lipids on tissue partitioning of HOCs.

420 **Enantiomeric Ratios and Metabolism of α-Hexachlorocyclohexane in Treated Rats and Cells.** Willett, K.L., Ulrich, E.M., Hites, R.A. Indiana University, Bloomington, IN; Caperell-Grant, A. Bigsby, R.M. Indiana University Medical School, Indianapolis IN. Technical hexachlorocyclohexane (HCH), an organochlorine pesticide, contains various HCH isomers including two enantiomers of α-HCH. While technical HCH contains enantiomers in a 1:1 racemic mixture, variable

enantiomeric ratios (ER) have been reported in environmental samples. This study investigated α-HCH metabolism and effects of the P450 inducer phenobarbital (PB) in treated rats and H411E rat hepatoma cells. In cells dosed with 10^{-6} M α-HCH for 24 or 48 hr, with or without PB pretreatment, no changes in cellular or media ERs were found. In tissues collected from male rats treated with 25 mg/kg α-HCH (1 or 24 hr) following PB pretreatment (75 mg/kg; 4 days) or control (no inducer) there were tissue specific differences in accumulation and ERs. No differences between treatments were seen in α-HCH concentrations at 1 hr in blood, fat, or brain tissues. In contrast, by 24 hr significantly higher concentrations of α-HCH were detected in all tissues from control rats; indicating PB significantly induced metabolism of α-HCH. Highest concentrations were in fat followed by brain and liver samples. ERs (+/- enantiomer) regardless of treatment regime were 1.19 ±0.62 and 1.24± 0.45 for blood and fat, respectively. However, in brain samples dramatic increases in ER were detected even by 1 hr (ER=3.0± 0.59). By 24 hr, ERs were 12.6 and 5.2 for PB and control brains, respectively. Therefore, α-HCH is differentially accumulated in rat tissues, ERs are tissue dependent, and PB induction has profound effects on both tissue accumulation and ERs. We are investigating whether + α-HCH enrichment in brain tissue is due to differential metabolism or blood-brain barrier interactions using rat as a model.

422 Bioassessment of Multiple Stressors - the Straight Poop. Munkittrick, K.R., Environment Canada, Burlington, Canada; Suter, G., Oak Ridge National Labs, TN; Antcliffe, B., Dept. Fisheries & Oceans, Vancouver, BC; Davis, W.R., US EPA, ORD/NHEERL, Narragansett, RI; Dyer, S., The Procter & Gamble Company, Cincinnati, OH; Gerritsen, J., Terta tech, Inc., Owings Mills, MD; Linder, G., Heron Works Farm, Salem, OR; Rankin, E., Ohio EPA, Columbus, OH. The bioassessment work group formed part of the Pellston Workshop on Multiple Stressors in September 1997 at Pellston MI. Our work group focused on the strategy and intent behind biological assessment and its relationship to ecological risk assessment for multiple stressors. The presentation describes a framework for bioassessment. While it need not be equated to ecological risk assessment, biological assessment shares many concepts with that process because the evaluation of valued ecological components (VECs) may represent an initial measure of adverse effects associated with a multiple stressor exposure. Following a description of the bioassessment framework, the relationships between biological assessment and ecological risk assessment were identified and characterized. These relationships focused primarily on the role of biological assessment in the analysis of risks associated with multiple stressors. The bioassessment can also play a role in developing cause-effect relationships to identify stressors associated with impairments in VECs. The group discussed planning and study design, biosurveys, determination of subnominal states, reference population approaches, analytical approaches, identification of potential causal relationships, and risk management, remediation and protection. The group concluded with discusisons of bioassessment linkages to stressor-based risk assessments and an overview of knowledge gaps and research needs.

423 Identifying Multiple Stressors in Ecological Risk Assessment. Klump*, J. V., U. Wisconsin-Milwaukee, Milwaukee, WI; Harris, H.J., U. of Wisconsin-Green Bay, Green Bay WI; Adams, W.J., Kennecott Copper, Magna UT; Cardwell, R., Parametrix, Kirkland WA; Fairbrother, A., ecological planning & toxicology, Corvallis OR; Ingersoll, C.G., USGS, Columbia MO; Power, M., U. Manitoba, Winnepeg MB; Reid, L.M., USDA Forest Service, Arcata CA. Understanding the relationship between environmental integrity and human health requires a synthesis of information and knowledge about the workings (structure and function) of a given ecosystem and the stressors (anthropogenic or "natural") that have damaged the integrity of the ecosystem. If the system in question exhibits (or will exhibit at some time in the future) behavior contrary to the long term persistence of the valued natural capital of the system, then it follows that there are stressors to be measured. To begin a risk assessment, it is critically important to clearly define the questions being asked and to document the formulation of the problem in such a way that the future readers will understand the reasons why particular system components were included or excluded. It is helpful to phrase the question in the general form of "what is the probability of change to valued parts of the ecosystem if a new set of stressors is imposed on the current system, given that the system already is stressed by natural and anthropogenic forces." Aspects that should be considered when assessing the risk of multiple stressors to ecosystem components or processes, include (1) formation of a diverse team of experts to develop a conceptual model of the system at risk, (2) development of the broad conceptual model, (3) identification of an inclusive list of candidate stressors, (4) evaluation of multiple iterations of the conceptual model to select and prioritize a short list of primary stressors and their potential interactions, and (5) establishment of peer review at all key points in the process.

424 Linking Stressors and Ecological Responses. Warren-Hicks, W.J.*, The Cadmus Group; Solomon, K.R.R., Canadian Centre for Toxicology; Gentile, J. H., University of Miami; Butcher, J., Tetra Tech; Rattner, B. A., Department of the Interior; Landis, W. G., Western Washington University; Wenger, R., University of Wisconsin - Green Bay. This paper will review the critical findings of the working group entitled *Linking Stressors and Ecological Responses*, as part of the Pellston Conference on Multiple Stressors held in September 1997, Pellston, MI. Broadening the traditional concept of risk assessment to address multiple stressors and multiple ecological effects requires new tools. These tools must accommodate a variety of stressors and a range of ecological responses. In the simplest case, a biological response may result from a single stressor. But that same response may result from another stressor, or even a combination of several stressors. Reciprocally, one stressor may link to (impact) only one ecological response or it may link to numerous responses across several assessment endpoints. To characterize ecosystem risk it is necessary to identify the linkages between chemical, physical, and/or biological stressors and ecological endpoints. The goal of a risk assessment is to describe these linkages in a precise quantitative manner. However, because of the complexities of the interactions between stressors and ecological effects, and, for that matter, among the stressors themselves, it is not always possible to develop quantitative stressor-response profiles. Nevertheless, it is important that a conceptual framework be employed to characterize the full array of linkages between stressors and ecological effects, including those linkages that can be described only in a qualitative manner. Thus, this paper commences with a conceptual representation of the problem of developing a linkage analysis for multiple stressors and responses. The remainder of this paper surveys a variety of methods for accomplishing the linkage analysis for multiple stressors.

425 Risk Characterization in Ecological Assessments for Multiple Stressors. deFur, P.L., Virginia Commonwealth Univ., Richmond, VA; S. Ferenc, ILSI, Washington DC; M. Dobbs, Bayer Corp., Stilwell, KS.; C. Grue, Washington Univ., Seattle, WA; S. Marcy, U.S. EPA, Anchorage AK; D. Moore, Cadmus Group, Ottawa, ON; R. Rolland, Tufts Univ., Boston, MA and R. Wentzel, U.S. EPA, Washington, DC. This workgroup addressed issues regarding risk characterization for multiple stressor situations; risk characterization being the final phase in a full risk assessment. The workgroup acknowledged that characterizing these risks includes many factors common to other situations: quantitative estimates of the risk and descriptions of the limitations, uncertainties and unknowns. But multiple stressor situations are often larger in scale, not uncommonly a watershed scale, and may include multiple species or properties of ecosystems that are not exhibited by single species or individuals (e.g. diversity, energy transfer). The workgroup used examples from Green Bay, WI, Waquoit Bay, MA and non-indigenous species introductions, in our deliberations. We recommended using a matrix approach to sort out the types and severity of stresses and consequences. We further recommended an iterative, or progressively more detailed approach to the characterization of multiple stress situations. The workgroup identified several issues of special concern for multiple stressors: effects not associated with known receptor responses; cascades of risks; temporal variations between (and within) exposures and responses; need for clarity in process. The quotient method was rejected as a usable tool to characterize these types of risks. Expressing risks from different sorts of stresses and integrating exposures and responses is especially challenging and new methods and tools are needed to advance this area of the field. Multidisciplinary teams have been particularly successful and should be routinely used in all multiple stressor cases.

426 **Managing Ecological Risks Posed by Multiple Stressors.** Stahl, R.G.*, DuPont, Wilmington, DE; Miller, J., USEPA, Washington, D.C.; Frederick, R., USEPA, Washington, D.C.; Courtemanch, D., Maine Dept Environmental Protection, Augusta, ME; Frissell, C., Univ of Montana, Polson, MT; Kaplan, M., New Jersey Dept Environmental Protection, Trenton, N.J.; Sappington, K., USEPA, Washington, D.C.; Zeeman, M., USEPA, Washington, D.C. An important component of environmental protection is managing risks to ecological resources, an activity made even more difficult when addressing risks from multiple stressors. This activity requires the risk manager to contend with significantly increased biological, ecological, social and jurisdictional complexity. In addition, the state of the science of ecological/biological assessment is such that assessors may not have adequate tools to provide the information needed for effective and scientifically supportable decisions. We describe some of the important tasks addressed by the risk manager/risk management team, their role in the assessment and decision making process, and recommendations for the type of information and a format that would be relevant to the management decision. Acknowledging that experienced risk assessors and managers may be very familiar with the concepts described within particular recommendations, the complexities faced in managing risks from multiple stressors warrants (or strongly argues for) consideration of these ideas in combination.

427 **Interaction Between Landscape Forms, the Impacts of Contaminants, and Other Stresses.** Anderson, M.S., and Landis, W.G.*, Institute of Environmental Toxicology and Chemistry, Huxley College, Western Washington University, Bellingham, WA. A common problem encountered in assessing the impacts of multiple stressors in ecological systems is discerning stressor effect from site-specific spatial and temporal heterogeneity. At issue is the question of whether it is the contaminant or the cleanup process that creates more of an impact in altering the patch dynamics of the intrinsic system. We have examined this problem by conducting a literature review and applying patch dynamics to our distance explicit patch dynamic models. The literature contains very few examples that incorporate contaminants into a landscape context. However, certain attributes of patch distribution are critical in determining recolonization after an impact. These attributes include distance from a source population, shape of the patches (total edge to area), and whether the patch is a core surrounded by satellite patches or a satellite itself. A contaminant, in addition to its direct toxicological effects, can also alter the patch dynamics of a landscape by acting as a block to migration and recolonization. Conversely, if the cleanup of a contaminant severely alters the type of habitat, the results may be long-term impacts upon landscape structure. We are using our patch dynamic models to explore each of these scenarios by adding contaminants to several patches and also altering the patch layout to simulate habitat alteration. Preliminary findings indicate a variety of dynamics can occur from seemingly simple alterations in population size, patch layout, stress distribution, and changes in migration distances. Contamination along with the effects of cleanup procedures need to be considered in the context of the landscape to predict successfully the impacts of each to the system.

428 **Watershed Enhancement Plan Development for Leading Creek after Dewatering of the Meigs 31 Coal Mine in Ohio.** R.J. Currie*, D.S. Cherry, J.E. Babendreier, H.A. Latimer, D.M. Johnson and J.H. Van Hassel. Virginia Tech, Blacksburg, Virginia, Ferrum College, Ferrum, Virginia and American Electric Power Service Corporation, Columbus, Ohio. Litigation over the dewatering of Southern Ohio Coal Company's Meigs #31 mine lead to the development of a proactive plan to evaluate the effects of the mine dewatering, recovery of the stream system and development of a watershed enhancement plan. Although natural processes have lead to recovery in Leading Creek from the impact of dewatering, watershed reconnaissance has shown significant problems relating to toxicity and habitat degradation in the watershed. Seventeen tributaries and ten mainstem stations have received monthly monitoring as point source discharges, including biological, chemical, toxicological, and hydrological sampling. Evaluation of habitat impairment has included habitat assessments, in-stream riparian surveys and land use analysis. The enhancement plan began with the ranking of the ten mainstem stations and seventeen tributary stations based on prioritization of impact parameters, including agricultural sedimentation, sedimentation from Abandoned Mined Lands (AMD), poor water quality and multiple toxicity endpoints. Mainstem and tributary rankings were supported by analyses generated by the Hydrological Simulation Program-FORTRAN (HSPF) used to predict flow and water quality conditions. Prioritization of enhancement activities was developed for each station based upon the above parameter inputs along with best professional judgement.

429 **A Framework for Ecological Risk Assessment of Multiple Activities.** Suter, G. W., Oak Ridge National Laboratory, Oak Ridge, TN. The standard framework for ecological risk assessment does not explicitly address multiple activities or indirect causation. Although this has not prevented its use for assessments of risks from multiple agents, the routine assessment of complex programs or of multiple agents acting on a site, watershed or region would be aided by use of a framework that is designed for that purpose. The framework proposed in this presentation is modular with respect to the individual activities, which makes the assessment more manageable and more efficient when the same activities are addressed in multiple programs or at multiple sites. It explicitly allows for analysis of indirect effects in terms of causal chains. It includes links to other risk assessments for which changes in ecological conditions are the hazardous agent. For example, changes in ecological condition may create risks to recreational uses, fisheries, or to the cultural resource values of a site. The framework also includes a standard approach to estimating the combined effects of the multiple agents acting on a receptor based on co-occurrence and the degree of mechanistic similarity of the modes of action. Finally, the framework includes an approach to developing conceptual models that is efficient yet mechanistic. This work was supported by a grant from the Strategic Environmental Research and Development Program.

430 **Physical and Chemical Factors Affecting the Aquatic Community in the Upper San Juan River Basin.** Allert* A.L., Environmental and Contaminants Research Center, Columbia, MO, Thorn, T.D., New Mexico State University, Las Cruces, NM, Finger, S.E., Environmental and Contaminants Research Center, Columbia, MO, Callahan, E., Environmental Statistics, Fountain City, WI, Steiner, R.L., New Mexico State University, Las Cruces, NM, Caldwell, C.A., New Mexico State University Cooperative Research Unit, Las Cruces, NM. An assessment of the macroinvertebrate community, contaminants, and water and habitat quality was conducted to determine whether these factors could be limiting populations of razorback suckers (*Xyrauchen texanus*) and Colorado squawfish (*Ptychocheilus lucius*). Metal concentrations in sediment, surface and porewaters were low, although significant differences were found in surface waters between sites. Evaluations using Toxic Unit (sediment) and Hazard Quotient (surface water) modeling indicated potential risk to the aquatic community from metals. Concentrations of 16 polycyclic aromatic hydrocarbons (PAHs) were also low in sediment, porewater and Semipermeable Membrane Devices. Sufficient solar radiation exists in the San Juan Basin to photoactivate PAHs. Evaluation of seasonal differences in solar radiation could be important in determining whether the aquatic community is at risk of increased exposure during low flow periods. No significant mortality in juvenile Colorado squawfish occurred during the *In situ* exposure. Site porewaters were not acutely toxic to *Ceriodaphnia dubia*, however reproduction differed significantly between sites. Macroinvertebrate community composition differed significantly between sites and sampling method. Use of an artificial substrate was effective and demonstrated the importance of habitat quality to invertebrate

community composition. Degradation of physical habitat, changes in water quality, and the presence of contaminants in the San Juan River may be preventing the recovery of the endangered fish populations.

431 Patterns of Liver Tumor Prevalence in Brown Bullhead from the Black River, Ohio, Before and After Remedial Dredging. Baumann, P.C., U.S.G.S., Columbus, OH; Since 1980 liver neoplasms in brown bullhead and polynuclear aromatic hydrocarbons (PAH) in sediment have been researched in a series of studies on the Black River near Lorain, Ohio. In the early 1980s the liver cancer prevalence in fish of age 3 and older was high, ranging from 22% to 39% of the adult population. These high cancer rates corresponded to high levels of PAHs in the sediment resulting from long-term releases by an upstream coking facility (USX). In 1983 this coking plant was closed, and by 1987 the PAH in sediment had declined by about two orders of magnitude. Coincidentally the tumor prevalence in 1987 was only about one-fourth of that in the early 1980s. In 1990, following a USEPA Consent Decree, the area having the most contaminated sediments were dredged. Neoplasm surveys in 1992 and 1993 found liver tumor frequencies in mature bullhead were as high or higher than in the early 1980s. However liver tumor incidence declined in 1994, especially among age 3 fish, where neoplasm incidence was zero. These age 3 fish were the first group sampled that were not present during the 1990 dredging. Limited data from 1995 and 1996 indicate that liver cancer prevalence has remained low (4% to 12%). These data are consistent with a hypothesis that the increase in tumor prevalence in 1992 and 1993 was caused by exposure to buried PAH-contaminated sediments released by the dredging. However this increase was restricted to the age groups present in the river during the year of the dredging, with fish hatched in later years having a reduced cancer prevalence.

432 Cytotoxic and Genotoxic Effects of Some Energetic Compounds Tested on Bacterial and Mammalian Cells *In vitro*. Lachance, B.*, Hawari, J., Robidoux, P. Y., Sunahara, G., Biotechnology Research Institute, National Research Council of Canada, Montreal, QC, Canada ; Ampleman, G., Thiboutot, S., Defense Research Establishment-Valcartier, Ministry of National Defense, Val-Belair, QC, Canada. Several energetic compounds present in explosive-contaminated soil and water samples, namely trinitrotoluene (TNT), trinitrobenzene (TNB), cyclotrimethylene-trinitramine (RDX), cyclotetramethylenetetranitramine (HMX), and various amino- and nitro- derivatives of toluene were evaluated. Their mutagenicity was studied using the Fluctuation test (FT), and the V79 mammalian cell assay. Their cytotoxicity was evaluated using the neutral red cytotoxicity test (NR) with V79 cells, and a growth-inhibition assay with human lymphoblast TK6 cells. In the V79 cell assay, only TNB was found more toxic than TNT. RDX and HMX were almost not toxic for both cell lines at their maximal solubility limit. The amino-dinitro and diamino-nitro metabolites of TNT were slightly less toxic than the parent compound in the NR assay. In contrast, the TK6 cells showed greater sensitivity to TAT, TNT and TNB, and were less sensitive to the metabolites. The results with the FT assay indicates that TNB, TNT and TNT metabolites are genotoxic, an effect which was sometimes decreased following the addition of S9. Amongst the agents tested, only 4-ADNT was found mutagenic using the V79 assay. Clearly, the bacterial genotoxicity assay is more sensitive to the effects of energetic compounds than are the mammalian V79 cells, and would then constitute a good monitoring tool for genotoxicity during a bioremediation process. The results of this study suggest that the obvious loss in cytotoxicity associated with the production of TNT metabolites is not accompanied by a simultaneous reduction of the mutagenic potential. Also, since at least one mammalian cell mutagen is produced by reduction of TNT, it follows that all of its breakdown products should be measured during bioremediation.

433 Comparative Analysis of Sediment Extracts from NOAA's Bioeffects Studies by the Biomarker, P450 RGS. Jack W. Anderson and Jennifer Jones, Columbia Analytical Services, and Jawed Hameedi and Edward Long, National Oceanic and Atmospheric Administration. Sediment samples collected from nine coastal and estuarine areas have been analyzed to determine response of a biomarker, P450 Reporter Gene System (RGS). This biomarker, using a transgenic cell line, detects the presence and potency of organic compounds that typically induce the CYP1A gene. Previous investigations have shown that sediment stations producing a response of 60 μg/g of benzo[a]pyrene equivalents (B[a]PEq) also exhibited a degraded community structure. RGS data from all study areas combined (527 samples) show that the mean response is 22.7 μg/g B[a]PEq, and the mean upper 99 % confidence interval is 32. We believe that sediments exhibiting B[a]PEq values of 32 to 60 are possibly contaminated to the extent that effects on benthic organisms may be observed, and those producing an RGS response of 60 and greater are likely to be toxic. The majority of the stations investigated produced responses below 32 μg/g of B(a)P equivalents. No samples from Biscayne Bay, Galveston Bay, Winyah Bay, or the small bays and lagoons of southern California produced responses greater than 39. However, samples producing induction over 60 μg/g B[a]PEq were: 50%, San Diego Bay; 16%, Delaware Bay; 8%, Sabine Lake; 4%, N. Puget Sound; and 3%, Charleston Harbor. A strong statistical correlation was observed between the RGS responses and high molecular weight PAH concentrations in the sediments from several of the study areas. Of the 2694 km^2 encompassed in the surveys, only 0.42 % of the area exhibited responses above 32 μg/g B[a]PEq, which represented 11.4 km^2. This assay is useful in identify high levels of toxic and carcinogenic compounds in the sediments and predicting their likely impact on the biological community.

434 Evaluation of the Genotoxicity of Anthropogenic Chemicals Using Transgenic Fish. Gallagher, K.*, National Institute of Environmental Health Sciences, Research Triangle Park, NC, J. Cline, Geo-Centers Inc., Ft. Detrick, MD, J.G. Burkhart, NIEHS. A transgenic model system has been used to evaluate the effects of mutagens in fish liver in *in vivo* studies. This model system uses a transgenic reporter, the bacteriophage ¯X174*am3*, integrated into the mummichog (*Fundulus heteroclitus*) genome. Mummichogs, present in saltwater marshes from Maine to Florida, frequently inhabit industrialized estuaries, and have been proposed as useful sentinels of environmental conditions. The ¯X transgene enables identification of DNA base substitutions at a single A:T pair in DNA recovered from exposed fish. This model system has been evaluated with both direct- acting and indirect mutagens. The effects of the direct-acting mutagen, *N*-ethyl-*N*-nitrosourea(ENU), on *in vivo* mutant frequency have been examined. Aqueous exposure to 100 mg/L ENU for one hour resulted in a 21-fold increase in mutant frequency in ¯X recovered from fish liver 42 days after exposure (six-fold, excluding one high outlier). We have also exposed transgenic fish to a model polycyclic aromatic hydrocarbon, 7,12-dimethylbenz[a]anthracene (DMBA), which must be metabolized to assume its full mutagenic potential. Animals were intraperitoneally injected with doses of 0, 1.9, 10 and 19 mg/kg DMBA in corn oil and were sacrificed after 30 days. The 19, 10 and 1.9 mg/kg doses of DMBA resulted in a seven-fold, five-fold and two-fold increase, respectively, in ¯X mutant frequency recovered from liver. Experiments are currently underway to examine the mutagenicity of natural sediment collected from a known polluted site in the Chesapeake Bay, relative to that of a known clean site, in order to determine if this transgenic model system would be useful in assessing the aquatic toxicity of natural sediments at sites with suspected anthropogenic contamination.

435 Spontaneous and Induced Mutations in λLIZ Transgenic Medaka. Winn, R. N.,* Norris, M.B., University of Georgia, Athens, GA. Transgenic animal models capable of measuring mutations are providing powerful new tools for rapidly assessing mutations *in vivo* following mutagen exposure. The general approach entails the introduction of a recombinant vector that carries a mutation target gene into the animal genome, and following exposure to a mutagenic agent, recovering the target gene via indicator bacteria to quantify the frequency of mutations. We describe the development and initial validation trials of transgenic medaka (*Oryzias latipes*) that carry multiple copies per cell of the bacteriophage λLIZ vector. Previously developed in rodent models, the λLIZ vector carries both the *lac I* and the *C-II* genes as target genes which are each analyzed by different mutation detection assay systems. Of fifty-five transgenic founder fish produced, 15 demonstrated germ-line transmission of the transgene ranging from 1to ~100 copies/haploid genome. Hemizygous and homozygous transgenic fish from high gene copy number lineages were examined with regards to the efficiency of vector recovery, spontaneous mutation frequency, and induced mutation frequency of the *lac I* and *C-II* mutation target genes following exposure to the model mutagen ENU. Procedures were developed to enhance the yield of high-concentration, high-molecular weight genomic DNA, which in turn facilitated successful packaging and highly efficient recovery of the λ vector from transgenic fish (3-8 X 10^5 phage/packaging reaction). The spontaneous mutation frequencies of the target genes in the fish were comparable to the range of frequencies observed in transgenic mice with the identical mutation targets (2-8 X 10^{-5}). The mutation spectra and the mutation frequencies detected by the *lacI* and the *C-II* genes following mutagen exposure were similar to one another, and to those observed in λLIZ mice following ENU exposure. These results indicate that the fundamental requirements for the transgenic fish models for mutation detection have been met and that these models may find a significant applications in genetic toxicity testing, with obvious environmental relevance to the aquatic systems.

436 Evolutionary Responses in Contaminant-Exposed Populations: Detection and Consequences. Diamond, S.A. U.S. EPA, Duluth, MN. The genetic consequences of contaminant exposure continue to receive attention from ecotoxicologists. Methods for detecting genetic differences among populations residing in different habitats or microhabitats have been adopted by toxicologists in an effort to discern whether contaminants act as a significant selection force in the environment. Two central hypotheses underlie these efforts: One is that unique allelic frequencies produced by contaminant selection can serve as a biomarker for otherwise indiscernible population-level contaminant effects; the second is that these genetic, contaminant-imposed evolutionary processes result in less robust (or adaptable) populations. These hypotheses are often accompanied by conflicting assumptions that heterozygotes are "more fit", yet alleles at affected loci can be lost by selection. An alternate (and perhaps more parsimonious) hypothesis is that genetic variability is lost by bottlenecking, and that the alleles lost are not specific to the contaminants present. The hypothesis that the relative robustness of populations adapted to contaminants has rarely been tested (excepting cases where the putative selecting stressor was used in comparative exposures). Several acute and chronic exposures were conducted to address several aspects of these hypotheses. Differential survival among allozyme genotypes was apparent in metal and PAH exposures, although results were inconsistent among exposures. The results of selection exposures varied for fluoranthene/UV exposures; acute selection resulted in a less tolerant population whereas chronic selection produced a more tolerant population. Effects on measures of robustness, including swimming capacity, growth, and egg production suggested that chronic selection did not have a general fitness consequence, while acute selection had very significant fitness consequences. These results, as well as selected, pertinent literature examples will be used to address several issues associated with evolutionary responses to contaminant exposure.

437 Recent Advances on Population Genetics Studies of Meiofauna Taxa Inhabiting Contaminated Estuaries in Southeast US. Schizas N.V.*, Street G.T., Gordos M., Chandler G.T., Coull B.C., University of South Carolina, Columbia, SC. A prerequisite for testing hypotheses on toxicant effects on the population genetic level of organisms is the knowledge of population structure of the species of interest. Because very little is known about the population genetics of meiofauna taxa over large geographic scales, it is important to first determine the systematic relationships among different populations of species of interest. The identification of discrete population units by phylogenetic analysis will facilitate the evaluation of toxicant stresses as a micro-evolutionary force. When population units are defined, specific hypotheses about individuals inhabiting creeks or bays with different contamination histories can be successfully addressed such as do sediment contaminants reduce the genetic diversity of benthic organisms. We have applied this approach to two meiobenthic species inhabiting contaminated estuaries in the southeast US: the copepod *Microarthridion littorale* and the foraminiferan *Ammonia beccarri.* The population structure of these taxa has been deciphered with concomitant examination of independently evolved mitochondrial and nuclear genes. On a large-scale study, we identified discrete populations of *M. littorale* that coincide with state geography, but apparently have no measurable gene flow among them. On a fine-scale study we measured genetic diversity and gene flow of both copepods and foraminifera in several creeks within the southeast US. Contrary to our expectations, genetic diversity was not correlated with the contamination history of the creeks we examined. Our conclusions are further supported by bioassays indicating absence of increased tolerance and an enhanced reproductive sensitivity to pollutants for copepods regardless of their creek of origin.

438 Dna Variation in Copepods (Diaptomus mississippiensis) and Alligators (Alligator Mississippiensis) Inhabiting Ponds Contaminated with Radionuclides. G. Street*, T. Glenn, and N. Schizas, University of South Carolina, Columbia SC, and B. Taylor, Savannah River Ecology Lab, Aiken SC. Extending risk assessment to the genetic level is an emerging, novel approach to aquatic toxicology. While interpretation of DNA data for purposes of contaminant risk assessment is still in its infancy, there are several advantages of this method over traditional bioassays. Relative to molecular genetic studies, measuring phenotypic effects (e.g. mortality, growth, and reproduction) while sometimes effective, can be time consuming and insensitive to chronic, low-level exposures. Of all the common anthropogenic toxicants, radionuclides have the greatest potential to directly affect DNA sequence variation. The US DOE's Savannah River Site (SRS) contains several reactor cooling ponds, some of which are contaminated with low levels of radionuclides, including cesium-137. The remediation of such sites is projected to be lengthy and expensive. We are involved in an ongoing study to measure variation in DNA of organisms inhabiting ponds and reservoirs within the SRS to ascertain whether increased mutation rates can be detected. We are targeting two organisms: an abundant, rapidly-reproducing planktonic crustacean near the bottom of the food chain (the calanoid copepod Diaptomus mississippiensis) and a slow-growing vertebrate at the top of the food chain (the American alligator, Alligator mississippiensis). DNA is extracted from individual organisms, amplified using PCR, and then either directly sequenced or screened for microsatellite variation. Preliminary results indicate a low degree of sequence divergence among alligators, and a high degree of sequence divergence among copepods. We will attempt to relate contaminant concentrations to variation in copepods, taking care to separate out the effects of historical biogeography (e.g. colonization and extinction of local ponds.) Relating genetic variation to contaminant concentrations should eventually provide a powerful new tool in the arsenal of risk assessors.

439 Measurement of DNA Damage, Growth, and Bioaccumulation in Infaunal Mussels Deployed at Stations in San Diego Bay. Steinert, S.A.* and Streib Montee, R., Computer Sciences Corporation, San Diego, CA. Maintaining DNA integrity is of paramount importance to all living things. In previous studies at sites in San Diego Bay DNA damage, growth and contaminant bioaccumulation were followed in *Mytilus edulis* deployed in the water column. DNA damage was found to be higher at sites with elevated sediment contaminant levels. In this study infaunal mussels, *Muscuslista senhousia*, were deployed in the sediment at these previously studied sites. The mussels were deployed for 32 days and growth, contaminant bioaccumulation and DNA damage, determined using the comet assay, were measured. In addition, a method incorporating ultraviolet light photoactivation was used as a diagnostic to determine the bioaccumulation of photoactive compounds.

Growth at all stations was significantly lower than that found at the reference site. Significantly higher levels of DNA damage were found in germ cells at 2 stations. The mussels at these stations bioaccumulated PAHs and metals at significantly higher concentrations, and the cells from mussels at these stations alone showed significant photo-induced DNA damage. Comparison of the results from this study and the previous one highlight contrasting differences in sensitivity to various contaminants for *Muscuslista senhousia* and *Mytilus edulis. Mytilus edulis* appears to be more sensitive to metals uptake and relatively insensitive to PAHs in the absence of sunlight. *Muscuslista senhousia* appears insensitive to metals and sensitive to PAHs, even in the absence of sunlight. In addition, monitoring DNA damage following ultraviolet light treatment shows promise as a simple means of determining the bioaccumulation of some photoactive compounds.

440 The Isolation and Identification of Staghorn Coral cDNAs Induced by Permethrin and Copper Exposure in Tropical Marine Environments. Morgan, M.B., and T.W. Snell, Georgia Institute of Technology, Atlanta, GA. Differential display PCR was used to identify gene fragments that could serve as inducible biomarkers of stress from either the insecticide permethrin or copper. Ten-centimeter branches of Staghorn coral *Acropora cervicornis* were exposed for 4 hours to 1μ permethrin/L or Cu^{2+} $25\mu g/L$. Several differentially expressed cDNAs representing new gene products produced in response to permethrin and copper exposure have been isolated. The differentially expressed gene fragments were reversed transcribed to produce cDNAs that were subsequently isolated, amplified, and developed into probes. Chemiluminescent detection was used to identify when a probe annealed to its corresponding mRNA in dot blots. Verification of probe specificity and the elimination of "false positives" were performed by Northern blot analysis. Several true toxicant-induced cDNAs were sequenced. These sequences were compared to genes of known function by performing a blast search in several databases. Differential display PCR can be used to identify toxicant-induced genes that can serve as molecular biomarkers for rapid toxicity assessment in Scieractinian corals.

441 Comparison of Acute Toxicity Responses of Glochidia (Unionidae) to Six Chemicals.
Milam, C.D.*, Arkansas State University, Jonesboro, AR; Farris, J.L., Arkansas State University, Jonesboro, AR; Dwyer, J.F., USGS, Columbia, MO; Hardesty, D.K. USGS, Columbia, MO. The applicability of using toxicity assessments to protect listed species must be carefully evaluated by determining a range of sensitivities for both standard test organisms and suitable surrogate species. *Utterbackia imbecilis* has been used as a surrogate to represent early lifestage responses of freshwater molluscs, but toxicity endpoints suggest that this single species may not be predictive for all listed species. Acute toxicity tests were used in this study to compare responses in glochida of three freshwater mussel species (*Leptodea fragilis, U. imbecilis* and *Lampsilis cardium*) and two standard test organisms (*Ceriodaphnia dubia* and *Daphnia magna*). Carbaryl, copper, 4-nonylphenol, pentachlorophenol, and permethrin were used in exposures to represent different chemical classes and modes of action. No single chemical provided consistently high or low toxicity estimates; however, carbaryl and 2,4-D were generally the least toxic for all species tested. *L. fragilis* and *D. magna* were most sensitive to copper (LC50s of 0.09 and 0.12 mg Cu/L, respecively), while *L. cardium* was more sensitive to permethrin than to all other chemicals. Calculated LC50s for *U. imbecilis* were generally greater than for other mussels and some standard organism responses. *C. dubia* and *D. magna* had higher LC50 estimates than mussels for pentachlorophenol, 4-nonylphenol, 2,4-D and permethrin. Average LC50 values for pentachlorophenol were lower for *C. dubia* (0.30 mg/L) than *L. cardium* (1.25 mg/L). These data suggest that some standard organism and surrogate responses to the range of chemicals used in this study were not protective of the range of sensitivities exhibited by freshwater mussels during this early lifestage.

442 Sublethal Effects of Aerial Exposure on Three Species of Unionid Mussels. Greseth, S.L.*, Rada, R.G., University of Wisconsin-La Crosse, La Crosse, WI; Waller, D.L., Bartsch, M.R., U.S. Geological Survey, La Crosse, WI; Cope, W.G., North Carolina State University, Raleigh, N.C. Freshwater mussels are emersed (exposed to air) during conservation activities, such as status surveys, relocations and reintroductions. Success of these activities depends upon the ability of mussels to survive emersion and burrow into the substrate. Sublethal effects of emersion were determined by changes in energy reserves of mussels. Three species of unionid mussels (*L. cardium, Q. pustulosa* and *E. dilatata*) were acclimated in water at 25 °C and exposed to five aerial temperatures (15 to 45 °C) for 15, 30 or 60 min. After emersion, mussels were returned to water at 25 °C. Burrowing and survival were observed for 14 d; mussels were then sacrificed and mantle tissue was analyzed for total lipid, carbohydrate, and protein content. All species demonstrated significant treatment effects on lipid content due to exposure temperatures. Carbohydrates within the mantle tissue of *Q. pustulosa* were significantly affected by aerial temperature and proteins within the mantle of *Q. pustulosa* were affected by duration of exposure. Proteins within the mantle tissue of *E. dilatata* were affected by aerial temperature. Most treatment effects were observed at the highest temperature of exposure indicating that conservation activities involving the emersion of mussels should be performed during air temperatures less than 45 °C.

443 Design and Evaluation of a Direct-Counting, Flow-Through Chamber to Test Toxicity to Developing Larval Clams, *M. mercenaria*,. LaBreche, T.M.C., Shepard, N.D., Lauth, J.R., Dietrich, A.M.; Virginia Tech, Civil and Environmental Engineering, Blacksburg, VA 24061-0246. A flow-through bioassay was designed to assess survival and development of free-swimming larval clams before metamorphosis into sedentary bottom dwellers. The design permitted: 1) retention of larvae < 70 μm, 2) continuous feeding, 3) direct microscopic counting without subsampling, 4) waste elimination, 5) sufficient sample for water quality analyses. An inner 30 mm petri dish bottom (organism dish) had two 1 cm sections removed and covered by 35 um screen. This was placed in 60 mm petri dish (catch dish) with a drainage tube inserted in the side. The organism dish was drip fed test solution from a reservoir via a 1 mm i.d. tube inserted through the catch dish top. Test solution filled then drained autonomously from the dishes; 10+ dish volumes per hour were exchanged at 1 L/day. Dead and live organisms were counted by removing the organism dish and placing on a microscope for inspection, photography, or videography, then returning the organism dish to the catch dish.
Triplicate tests with daily counting were conducted starting with 6 day, free-swimming *M. mercenaria*. $CuNO_3$ solutions were prepared in 26.5 ppth salinity (Instant Ocean®); larvae were maintained at 24°C and fed *Isochrysis galbana*. After 72 hours, nearly all the surviving clams had set. This design exhibited copper toxicity for larval *M. mercenaria* at similar concentrations as determined by Calabrese, *et al.*, 1977 who reported that 16.4 ug/L added copper was the 8-10 day LC_{50} in seawater containing 13.4 ug/L Cu.

Copper Conc. ug/L	% Survival,		
	24 hours	48 hours	72 hours
0	61	40	31
7.8	49	27	19
31.2	38	17	17

Removal of Aqueous Copper

444 The Sensitivity of the Juvenile Clam *(Mercenaria mercenaria)* **to Selected Toxicants Following Sediment Exposure.** Chung, K.W., Fulton, M.H., Scott G.I., NOAA/NOS, Charleston, SC. Littleneck clams *(Mercenaria mercenaria)* are marine filter-feeding, infaunal mollusks with great commercial importance along the East coast of the US. Laboratory sediment bioassays were conducted to determine the sensitivity of juvenile clams to three model contaminants (DDT, cadmium, and fluoranthene). The goal of this research was to evaluate the sensitivity of this test species as compared to that of other benthic and epibenthic organisms. Toxicity tests were conducted in 600 ml beakers containing 100 ml of sieved spiked sediment and 300 ml of 20 μm filtered seawater. Sediments were press sieved through a 212 μm mesh screen and spiked with the appropriate contaminant 24-h before start of the assay. The sediment was allowed to settle under aeration for 24-h before the addition of fifty clams per beaker. Clams were between 212 μm and 350 μm. Bioassays were conducted in environmental chambers at 20^{0}C and 30 ppt salinity with five replicates for each contaminant concentration and the control. After ten days, clam mortality was determined for each replicate. The mean LC5Os for the 10 day sediment exposures were <2.5 mg/kg for cadmium, <0.8 mg/kg for fluoranthene and 5.8 mg/kg for DDT. The LC5O for the juvenile clam was similar to the ERL (0.6 mg/kg) reported by Long et al. (1995) for fluoranthene. Likewise, the LC5O for cadmium in this species was quite similar to the ERL (1.2 mg/kg) value. Conversely, the LC5O for DDT in this species was considerably higher than the reported ERL value (0.0016 mg/kg). These findings suggest that the juvenile clam is one of the more sensitive species to a variety of toxicants and may be a valuable indicator for potential sediment toxicity.

445 Biomarker Studies with Juvenile Oysters Deployed ***In-Situ.*** Ringwood, A. H.*, Marine Resources Research Institute, Charleston, SC; Conners, D. E., Medical University of South Carolina, 171 Ashley Avenue, Charleston, SC; Keppler, C. J. Marine Resources Research Institute, Charleston, SC. Hatchery-reared juvenile oysters (*Crassostrea virginica*) were deployed *in situ* at estuarine sites that ranged from reference to highly degraded by urban/industrial pollution. A suite of cellular responses (lysosomal destabilization, glutathione concentrations, lipid peroxidation, heat shock proteins) were analyzed. Adverse effects on lysosomal destabilization were observed at all but one potentially degraded site. Glutathione depletion was observed at many of the contaminated sites and at one of the reference sites. Significant lipid peroxidation and increases in heat shock proteins were observed in oysters deployed at some contaminated sites and one reference site. Significant correlations between sediment contaminants and lysosomal destabilization and glutathione concentrations were observed. Although elevated lipid peroxidation products and heat shock proteins were observed more commonly at contaminated sites, there were poor correlations with contaminants. We present some evidence that heat shock proteins and lipid peroxidation may be related to dissolved oxygen stress. We hypothesize that lysosomal and glutathione responses may be used to identify significant perturbations associated primarily with contaminant stress, and that another suite of responses (heat shock proteins and lipid peroxidation) may be valuable indicators of oxidative stress. Alterations in cellular parameters represent primary level responses to environmental insults that could translate into reduced fitness, and eventually culminate into more extensive effects on community integrity. Cellular responses should be developed that can function as valuable early warning indicators of potential adverse effects, preferably before the effects are irreversible.

446 An Evaluation of Caged Mussels as a Monitoring Tool for Deepwater Effluents. Salazar, M.H., Applied Biomonitoring, Kirkland, WA; Salazar, S.M., EVS Consultants, Seattle, WA. A caged mussel pilot study was conducted between February and April, 1997, in Port Valdez, Alaska. The purpose of this study was to determine the feasibility of using transplanted mussels to monitor effluent from the Alyeska Ballast Water Treatment Facility (BWTF) at a depth of 70 meters where mussels are not normally found. At total of 2100 bay mussels (*Mytilus trossulus*) were transplanted from the intertidal zone in Anderson Bay to seven stations in the vicinity of the BWTF effluent diffuser for a period of 56 days. Bioaccumulation of polycyclic aromatic hydrocarbons (PAHs) was used to characterize potential chemical exposure. It was intended that growth of mussel shells and tissues would be used to characterize associated biological effects, but there was not a clear relationship between the effluent and mussel growth. Statistically significant differences in mussel growth were found among stations, and between stations inside the BWTF mixing zone and those outside it. Variability in data precluded the same statistical comparisons among stations and zones for mussel tissue chemistry. Nevertheless, the chemistry data showed a gradient of decreasing total PAH concentrations with increasing distance from the diffuser. The pilot study was successful because the objectives were met. Test mussels exceeded pre-determined survival and growth criteria and accumulated target chemicals in their tissues. The results demonstrate the feasibility of using caged bivalves for monitoring the effluent discharged by the BWTF. New information was provided regarding bioavailability, pathways of exposure, and potential exposure to PAHs. Results of the pilot study indicate that the caged bivalve methodology could be used to answer longstanding questions about the potential impact of the BWTF on Port Valdez.

447 Chambering in the Pacific Oyster, ***Crassotrea gigas*****, and its Relationship to Tributyltin Concentrations.** Brancato, M.S.*, Cardwell, R.D., Tear, L., McKay, S., Parametrix, Inc., Kirkland, Washington. Various field and laboratory studies have documented a relationship between tributyltin (TBT) exposure and the occurrence of growth abnormalities, including chambering and shell thickening, in *C. gigas*. The laboratory and field studies conducted in the 1980s presented exposure regimes where TBT water concentrations were many times higher than reported mean concentrations in U.S. waters. Mean water column TBT concentrations have been below the U.S. EPA's chronic marine water quality criterion of 10 ng/L for the past five years in the saltwater regions monitored, except commercial sites in Galveston Bay, TX, and marinas in Puget Sound, WA. Through an ongoing field study, we have determined that although the incidence of chambering was significantly correlated with TBT residue concentrations (especially tissue residues), the strength of the correlation was time and age dependent. Valve thickness was also significantly correlated with TBT residue concentrations in seedlings, but not in the one or two year olds. Interestingly, condition and weight showed no significant relationship with TBT residue concentrations or number of chambers. In other words, despite increases in the number of chambers and valve thickness, neither the weight nor condition of the oysters declined from that observed in the baseline population or from those transplanted to fish/shellfish areas. This indicates that TBT may be eliciting a response, but perhaps not an ecologically significant effect.

448 Intraspecific Differences in Zinc Accumulation in the Littoral Gastropod ***Littorina saxatilis*****.** Webb, S.F., Manchester University, UK. Intraspecific differences in zinc accumulation in the rough periwinkle was investigated in animals from 10 sites on the Isle of Man (Irish Sea). Metal handling ability was assessed in order to test the hypothesis that differences in prior metal exposure as manifested in zinc body burdens may also be reflected in differential metal accumulation. Observed zinc body burdens ranged from 43 to 155 μg/g dry weight. Elevated concentrations at certain sites (Peel & Laxey) are a consequence of historical anthropogenic metalliferous contamination. Animals (shell height 10 2mm) were identically exposed to 10 μg/litre added zinc-65 at 10 1 °C for 20 days. They were sampled at 5 day intervals and the levels of zinc-65 determined and expressed as μg/g dry weight (n = 351). Maximal uptake was $\leq$ 40 μg/g with a tendency for zinc-65 levels to *'plateau'* at the end of the experiment. Significant correlations ($p<0.05$) were established between zinc uptake and (i) length of exposure ($r = 0.63$), (ii) body weight ($r = -0.34$) and (iii) site zinc contamination ($r = -0.16$). A multiple regression of log transformed zinc uptake with components for length of exposure, body weight and site zinc contamination yielded an $R^2 = 0.58$. Overall, zinc uptake was significantly less in populations collected from contaminated sites (i.e., those with higher initial zinc burdens). Zinc uptake in animals from the cleanest site (Ramsey) was approximately 25% greater than in animals from the most contaminated site (Laxey) when estimated for average sized individuals (10 mg) after 20 days using the regression equation (NB: no significant correlation between animal weight and site zinc contamination). Results indicate that previously observed tolerance to experimental zinc exposure by populations from chronically contaminated sites could be due to reduced uptake.

449 Organochlorine and Trace Element Concentrations in Sediment and Mussel Tissues from the Lowermost Tennessee River and Kentucky Lake. Loganathan, B.G.*, Neale, J.R., Ratliff, J., Sickel, J., Owen, D.A. Murray State University, Murray, KY. Surface sediments and mussel tissue samples from the lowermost Tennessee River (Kentucky Dam Tailwater) and Kentucky Lake were analyzed for organochlorine compounds and selected trace elements in order to understand the status of environmental contamination by these toxic chemicals. The sediment and mussel tissue samples were freeze dried, Soxhlet extracted, cleaned-up and analyzed for organochlorine compounds using capillary column gas chromatography with electron capture detection (GC-ECD). Trace elements were measured using inductively coupled plasma atomic emission spectrometry (ICP-AES). Total PCB concentrations ranged from detection limit (<1.0 ng g^{-1}) to 26.36 ng g^{-1} dry wt. in sediments and 18.94 to 85.05 ng g^{1} dry wt. in mussel tissue homogenates. The greatest concentration of total PCBs was recorded in the samples (both sediment and mussels) collected from site at 17.7 TRM (Tennessee River Mile). Among the chlorinated pesticides, hexachlorobenzene (HCB) was consistently detected in almost all of the sediment and mussel tissues with highest concentrations (2.32 ng g^{-1} dry wt. in sediments and over 500 ng g^{-1} d wt. in tissues) at the same site (17.7 TRM). Other pesticides detected were DDTs and chlordane compounds, and the concentrations were detected at a few ng g^{-1}. Inorganic element concentrations in sediment and mussels were ranged from $\mu g\ g^{-1}$ to mg g^{-1} and the concentration pattern followed the order of Mn > Zn > Sr > Cu. The results give credence to infer that (i) Organochlorine contamination is relatively higher in the lowermost Tennessee River than in Kentucky Lake (ii) High levels (10^3 ppm or above) of Mn in sediments and mussel tissues has been well established as a "specific" indicator of Tennessee River and Kentucky Lake origin of such samples and (iii) Negative correlation of Sediment Mn and mussel tissue Mn suggested that mussels do not obtain significant amounts of Mn from sediments.

450 Scale Dependence of *Corbicula fluminea* Growth Rates Determined *In situ*. J.E. Babendreier*, D.L. Gallagher, Department of Environmental Engineering, D.S. Cherry, R.J. Currie, H.A. Latimer; Department of Biology, Virginia Tech, Blacksburg, VA. *in situ* toxicity testing using *C. fluminea* (Asian clam) has been identified as a tool for conducting ecological risk assessment of active and abandoned mined lands (AML). As part of an assessment of the Leading Creek Watershed, Ohio, the dependency of *C. fluminea* growth rates on stream flow rates and spatial scale was evaluated. The network design at Leading Creek included 10 mainstem sites, 17 tributary sites, and 3 reference sites. The creek's mainstem is 55 km draining 39,000 ha. Four *in situ* monitoring events using *C. fluminea* were conducted in June and August of 1996 and 1997, respectively. At each station, 25 juvenile clams (9 to 12 mm) were measured and marked, and five clams each placed in five separate pecan bags instream. The flow-through mesh bags where retrieved after 35 days and growth and survivorship determined. Along with physical and chemical monitoring, flow and water temperature were also monitored. The clam test events were each conducted under variable flow conditions ranging from bankfull to low flow. Sites were classified as impacted or non-impacted based on independent environmental data. A strong dependency was found between *C. fluminea* and spatial scale but not flow rate measured during the test conditions. A Monod-type model for predicting non-impacted growth rates as a function of drainage area was successfully fit to the data (maximum growth = 0.07 mm/day, K_s = 2700 hectare @ 20 C°, R^2 = 0.7). Consideration of drainage area in watershed-scale ecological risk assessments is thus required if *C. fluminea* tests are to be used to identify potentially impacted sites.

451 Weather-related Amphibian Activity Patterns and Water Toxicity: Comparative Risks of Exposure. Shipman, P.A.*, Bruner, M.A., Rao, M.N., Dumont, J.N., and Bantle, J.A. Department of Zoology, Oklahoma State University, OK. We examined the correlations between local weather parameters and amphibian activity patterns at a closed municipal landfill in Norman, Oklahoma, and at a nearby reference site. To monitor daily activity patterns over the three-year study, we captured animals using drift fence arrays with screen funnel traps and drop cans. Daily weather parameters, mean air temperature, minimum air temperature, cumulative rainfall, mean humidity, minimum humidity, mean wind speed, and solar radiation, were measured using on-site weather stations (Oklahoma Mesonet), and obtained via the Internet. We used a direct gradient analysis, canonical correspondence analysis (CCA), to show species and weather parameter relationships. Amphibian activity patterns, as expected, were positively correlated with rainfall, relative humidity, and air temperature and negatively correlated with wind speed and solar radiation, but not equally among species. Weather-related fluctuations in water toxicity at the landfill were previously characterized using Frog Embryo Teratogenesis Assay-*Xenopus* (FETAX). We use CCA scores to compare and evaluate the relative risks of exposure for amphibian species to weather-related changes in water toxicity.

452 Ecotoxicity and Environmental Conditions at a Closed Municipal Landfill in Central Oklahoma. Rao, M.N.*, Bruner, M.A., Dumont, J.N., and Bantle, J.A., Department of Zoology, Oklahoma State University, Stillwater, OK-74078. As part of our overall goal to investigate the possible environmental stressors that might be responsible for declining amphibian populations, we examined water toxicity and affects of environmental conditions. The study site is a closed municipal landfill in Norman, Oklahoma and is located on the flood plain of the Canadian River. Ground and surface water toxicity at the landfill was characterized using FETAX (Frog Embryo Teratogenesis Assay - Xenopus, a developmental toxicity assay). A high temporal resolution weather station (Oklahoma Mesonet) installed at the site provided the weather data. Surface water samples collected at least at monthly intervals (August 1997 to September 1998) were tested using FETAX and the results were correlated with the weather conditions existing around the sampling times. A significant correlation was observed between embryo mortality and weather conditions three days prior to surface water sampling. While cumulative rain and relative humidity showed negative correlations with mortality, solar radiation and net radiation showed positive correlations. However, weather conditions during days 4-7 preceding sampling showed no significant correlations. Ground water sampled from a single monitoring well showed 100% mortality throughout the sampling regime. These groundwater samples were subjected to Toxicity Identification and Evaluation (TIE) analysis. Mortality, and malformations were significantly reduced with the zeolite (removes ammonia) and mixed bed fractions (removes ionic constituents). Other fractions obtained following passage through activated carbon, C18 column, 0.2μ filtration, and aeration showed no significant reduction in toxicity.

453 Mortality, Growth, and Teratogenesis in Rana Spp. Exposed to Ambient Ultraviolet Light. Bruner, M.A.*, Shipman, P.A., Yoshioka, J.H., Dumont, J.N., and Bantle, J.A., Department of Zoology, Oklahoma State University, OK. We assessed the effects of ambient UV light exposure on native species of anurans during their normal breeding season in North Central Oklahoma. Eggs were collected from the field, sorted in the lab, and distributed equally among replicates of four different filter treatments (open, acetate, mylar, and shaded) of ambient UV light in outdoor experimental tubs. To dampen possible temperature differences among filter treatments, the experimental tubs were placed in water baths in 2000-liter artificial pools. Maximum-minimum recording thermometers were placed randomly in each treatment (one per treatment). Control replicates were also maintained in the lab for each experiment. The experiments began when the eggs were in late blastula stage and were terminated after the embryos had achieved stage 25 of development (nine days). Embryos, water levels, and water temperatures in each treatment were checked daily. We used a Biospherics, Inc. GUV radiometer to record daily ambient UV light exposure. Four endpoints (hatching failure, mortality, growth, and teratogenesis) were compared between filter treatments. Thus far, we have found no significant differences attributable to filter treatment.

454 Field and Laboratory Investigations of Amphibians within a Wetland Contaminated with Organochlorines. Sparling, D.W.*, USGS Patuxent Wildlife Research Center, Pinkney, A.E. USFWS Chesapeake Bay Field Office. A wetland adjacent to the Old Landfill, a hazardous waste site on the Marine Corps Combat Development Command, Quantico, was found to contain sediments contaminated with PCBs and DDT derivatives. As part of the ecological risk assessment for this site, we conducted field and laboratory investigations to determine if the contaminant burdens in the wetland could have adverse effects on amphibians. Field investigations showed that bullfrogs (*Rana catesbeiana*) and green frogs (*Rana clamitans*) inhabited the area and that at least green frogs bred there. Liver/body weight ratios were significantly smaller in green frog tadpoles collected from the wetland compared to an in-coming stream and to a reference wetland located in the nearby Mason Neck National Wildlife Refuge. In a 30-day bioassay employing green frog tadpoles and both unspiked and spiked sediments from sites within the wetland, no differences were detected in mortality or growth rates across treatments, but liver/body weight ratios were smaller in tadpoles from contaminated treatments. In a second bioassay with bullfrog tadpoles there was a significant difference in mortality among treatments with a greater frequency of mortality in spiked sediments than in controls. Three of nine bullfrog tadpoles that metamorphosed during the bioassay had ovotestes. Gonadal histology and residue analyses are being investigated further and will be reported.

455 Sublethal Effects of Coal Ash on Morphology, Behavior, and Physiological Energetics in Larval Bullfrogs. Rowe, C.L.*, University of Puerto Rico, San Juan, PR; Congdon, J.D., Savannah River Ecology Lab., Aiken, SC. Multiple characteristics of larval bullfrogs were modified by sublethal exposure to trace element-rich coal ash effluent. Bullfrogs inhabiting or transplanted into a polluted site had elevated whole-body concentrations of As, Cd, Cr, Cu, and Se compared to individuals in reference sites, and experienced sublethal effects on oral morphology, predator-escape behaviors, and expenditure of energy for maintenance. About 90 % of tadpoles collected from the polluted site had abnormalities in the oral region; such abnormalities were induced in 98 % of individuals transplanted to the polluted site as embryos. Presence of the abnormalities prevented effective grazing of periphyton, leading to reduced rates of growth in the laboratory when periphyton was a primary food source. Additionally, tadpoles collected from the polluted site were less responsive to prodding and had reduced swimming speeds compared to tadpoles from a reference site. Such modified behaviors may explain an increased susceptibility to predation in tadpoles raised in the polluted site compared to those raised in an unpolluted area. Finally, tadpoles collected from, or transplanted into, the polluted site had elevated allocation of energy to maintenance compared to those from unpolluted areas, after as little as 25 d exposure. The elevated maintenance expenditures in coal ash-exposed individuals likely reduces the net energy available for other energy-demanding processes. Thus, chronic exposure to coal ash sublethally modifies multiple systems which can have direct and indirect ramifications at higher levels of organization.

456 Elevated Trace Element Concentrations and Maintenance Costs in Banded Water Snakes, *Nerodia fasciata*, Exposed to Coal Combustion Wastes. Hopkins, W.A.*, Savannah River Ecology Laboratory, Aiken, South Carolina; Rowe, C.L., University of Puerto Rico, San Juan, PR; Congdon, J.D., Savannah River Ecology Laboratory, Aiken, South Carolina. Coal combustion waste is a ubiquitous form of global pollution that poses a significant threat to the health of wildlife. We currently are examining the sublethal impact of coal combustion wastes on banded water snakes, *Nerodia fasciata.* Water snakes inhabiting a coal ash disposal facility have significantly higher hepatic concentrations of As, Cd, and Se than conspecifics from a nearby reference site. To our knowledge, the levels of As and Se (mean = 134 and 142 ppm, respectively) in water snakes from the polluted site are the highest ever documented in a reptile species. In addition to being exposed to contaminants in the water and sediments, snakes in the polluted site are exposed to contaminants by ingesting prey items that have elevated whole body concentrations of As, Cd and Se. Snakes from the polluted site exhibited a mean standard metabolic rate (SMR) 32% higher than the mean SMR of snakes from the reference site. As a result, snakes from the polluted site appear to have increased allocation of energy to maintenance and therefore should have less energy available for growth, reproduction, and storage. Our findings are consistent with the physiological responses recently documented in other organisms from the polluted site. We hypothesize that long-term exposure to coal ash-derived trace elements and the resultant accumulation of some elements are responsible for the observed increases in maintenance costs.

457 Exposure and Effects of Pesticides and Toxic Elements in Wetland Reptiles in East-central Texas. Clark, D.R., Jr.*, U.S. GS, Texas A&M University, College Station, TX; Bickham, J.W., Texas A&M University, College Station, TX; Baker, D.L., U.S. FWS, Olympia, WA; Cowman, D.F., U.S. GS, Texas A&M University, College Station, TX. Blood samples taken from wild-caught diamond-backed water snakes (DBWS, *Nerodia rhombifer*), blotched water snakes (BWS, *Nerodia erythrogaster*), cottonmouths (CM, *Agkistrodon piscivorous*), and red-eared sliders (RES, *Chrysemys scripta*) were analyzed for organochlorine residues, toxic elements, acetylcholinesterase (ChE) levels, and genetic damage. Contaminated sites included a wooded slough with drainage from adjacent cotton and corn fields and an urban lake that received direct flow from another lake used as a dump for arsenic (As) compounds resulting during 53 years of manufacture of cotton defoliants. DDE at the agricultural site reached up to 3.0 ppm in whole blood of a DBWS. This much DDE in some bird species threatens reproduction. DDE residues were higher in both CMs and DBWSs than in RESs. In RESs at the agricultural site, mean ChE decreased with total mass (age) in both sexes, but the relationships were different with females having higher ChE than males of similar mass. ChE in RESs appeared to be continually depressed at the agricultural site. Mean ChE levels of RESs were less than thosse of DBWSs. Sites did not differ (significantly) in mean loads of toxic elements; however, in RESs means were greatest for all four measurable elements (copper Cu, mercury Hg, selenium Se, zinc Zn) at the agricultural site and amounts in DBWSs were greatest for three of the four (Cu, Hg, Se) at the agricultural site. Species differed in element loads at the agricultural site. Blood samples from the As-contaminated site contained no measurable As. There was no evidence of genetic damage.

458 Metals in Amphibians and Reptiles: An Ecological Risk Assessment View. Grillitsch, B. *, University of Veterinary Medicine, Vienna, Austria; Linder, G., Heron Works, Brooks OR. Ecosystem contamination from metals due to human activities is a matter of concern at a worldwide scale. Among vertebrate wildlife, ecotoxicology of metals in fish, birds, and mammals has been comparatively and extensively surveyed. In contrast, the assessment of exposure and effects of metals and their compounds in amphibians and reptiles has received very little attention. This presentation is based on an extensive literature survey on the ecotoxicology of metals in theses two vertebrate classes, and evaluates the pertinent information available with respect to general trends of toxicological kinetics and dynamics, overall research deficiencies, and priority research needs. Key findings of this review are: 1) metals may be considered potentially hazardous to free-ranging amphibians and reptiles with respect to environmental exposure, bioavailability and adverse effects; 2) toxicokinetics and toxicodynamics seem to follow general vertebrate trends in many respects; 3) there is indication for higher taxonomic level differences in metal metabolism and regulatory capacity; 4) conclusiveness of published research is highly limited due to general technical and methodical deficiencies; 5) for exposure and effect assessment, there is a significant lack of studies addressing 5.1) long-term low level exposure, 5.2) critical life stages and functional complexes; 5.3) biomarkers; 5.4) abiotic and biotic, exogenous and endogenous potentially interacting factors, 5.6) regulatory capacity with respect to acclimation, adaptation, and stress phases; hence, 6) there is too little information for meaningful ecological risk assessment particularly in terms of 6.1) interpretation of the ecotoxicological significance of metal tissue concentrations in wildlife; and 6.2) extrapolation among sites, taxa and levels of biological integration. In essence, these deficiencies as well as the resultant priority research needs seem to fall well within the trends generally recognized in ecological risk assessment. For amphibians and reptiles however, the inherent problems appear more impressive than in other vertebrates.

459 Comparative Toxicology and Risk Assessment of Amphibians. Westerman, A.G., Kentucky Department For Environmental Protection, Frankfort, KY; Kercher, M.D. and Birge, W.J.*, University of Kentucky, Lexington, KY. Effects of pollution on amphibian populations are difficult to quantify with the same degree of precision as for fish or macroinvertebrates. Due to wide variations in habitat, distribution and other factors, multimetric programs are less applicable. In addition, avenues of toxicant exposure may differ among amphibians. Assimilation via skin in anurans is proposed as a primary route of exposure. Another limiting factor in risk assessment for numerous amphibian species is the difficulty in obtaining sufficient numbers of organisms/eggs for toxicity testing. In this study, we propose to 1) review ecological assessment procedures; 2) identify chemical body burden thresholds for reproductive dysfunction; 3) evaluate the comparative toxicology of amphibians; and 4) develop Tolerance Indices (TI) for selected amphibian species versus Benchmark species (*e.g.* S_{LC50}/B_{LC50}; S_{LC10}/B_{LC10}). The comparative data base for amphibians included 187 toxicity tests with 34 metals and 19 organic compounds for up to 14 species representing five families, and toxicity values were determined with the embryolarval survival and teratogenicity test (Method 1001.0; *Federal Register*, 1995). Gonadal mercury residues of 0.5 μg/g correlated with reproductive dysfunction in certain species. Using rainbow trout as the benchmark, the median TIs for metals to the narrowmouth toad and atrazine to the American toad were 0.14-0.23 and 120, respectively. Median TIs for leopard frog versus rainbow trout were 0.2, 0.13, 0.44, 3.2, and 8.8 for dioctyl phthalate, phenol, benzene, Aroclor 1254, and atrazine, respectively. Other benchmarks included the South African clawed frog and the fathead minnow. The TI indicates greater species sensitivity (<1) or greater tolerance (>1) compared with established benchmark or surrogate species and, among other applications, can be used in preliminary risk assessments where more comprehensive toxicity data are lacking.

460 The Use of Reptiles in the Ecological Risk Assessment Process. Meyers-Schöne, L.*, IT Corporation, Albuquerque, NM. This paper reviews the discipline of ecological risk assessment and addresses how reptiles may be incorporated into the process to assess chemical and radiological impacts to reptiles in terrestrial and aquatic environments. In the evaluation of potential risks to the environment, certain species, or key receptors, within the ecosystem in question are selected as indicators of "environmental health". Reptiles are commonly used as biological monitors of chemical and radiological contamination, but are rarely used as ecological receptors in screening or definitive ecological risk assessments. Although reptiles generally meet most criteria for a good receptor (e.g., likelihood of exposure to the stressor, key component within a specific trophic level, and designation as protected or of recreational/commercial value for some species), the lack of sufficient toxicity information specific to reptiles renders utilization of this class of vertebrates in a risk assessment often difficult. A review will be presented on useful information obtained through biological monitoring studies that can be applied to retrospective ecological risk assessments that include reptiles. Examples will also be presented of studies where reptiles were key components in the risk assessment process. Finally, recommendations will be made of further areas of research needed to facilitate the use of reptiles in the ecological risk assessment process.

461 Water Issues Affecting the Florida Everglades. Jones, R.D.*, Florida International University, Miami, FL. The Florida Everglades are a unique, historically oligotrophic ecosystem threatened by the effects of pollution from agricultural and urban runoff. In addition, the federal Central and South Florida Flood Control Project (C&SF) has sectioned the historic Everglades system with a series of canals and levees to control water for urban and agricultural development. This has resulted in pronounced hydrologic modification to the natural system. This paper will describe the historic system and examine the effects of the manmade modifications, including pollutants and hydrology. Particular emphasis will be given to nutrient enrichment, mercury and hypersalinity in Florida Bay.

462 South Florida Ambient Pesticide Monitoring Program. Pfeuffer, R. J. South Florida Water Management District, West Palm Beach FL. The South Florida Water Management District (District) is an agency of the state created to manage surface and ground water quantity and quality in 16 counties in South Florida. Surface water and sediment have been sampled at various frequencies and locations in the District's 1400-mile system of canals since 1984, when the pesticide monitoring program was formalized. The program has evolved into over 30 sites with the surface water sampled quarterly and the sediment semi-annually for over 65 pesticides and degradation products. Selected sites have sediment monitoring annually. The most common pesticides detected in surface water are herbicide compounds, while DDE and DDD predominate in sediment samples. As a consequence of atrazine and ametryn being most often detected in surface waters receiving runoff from the 700,000-acre Everglades Agricultural Area, the agency has now included these compounds in its regulation program. Some exceedances of state surface water quality standards have occurred in certain basins. The temporal magnitude of the number of detections was determined for both the water and sediment.

463 Results of a Survey of Contaminants in Sediment, Water, and Fish from the Lower C-111 Canal and Selected Tributaries to Northeastern Florida Bay. Goodman, L.R., J.M. Macauley*, and T.H. Roush U.S. Environmental Protection Agency, Gulf Breeze, FL. A survey was conducted to determine contaminant concentrations in sediment, water, and fishes from the C-111 canal system and Northeastern Florida Bay during 1997. Twenty-seven sites were visited and samples collected during the spring period of low water flow and agricultural pesticide application. These were contrasted with samples collected during the fall period of high rainfall. Samples were analyzed for organochlorine pesticides using GC and GC/MS protocols, for total mercury using ICP, and for methyl mercury using ethylation GC separation and detection with atomic fluorescence. Concentrations of endosulfan I from 9.8 to 76 ng/ℓ were measured in water samples from four canal sites during March, but none were detected during the September sampling. Sediment concentrations for endosulfan I for 9 sites ranged from 0.88 to 5.6 ng/g. The endosulfan breakdown products, endosulfan II & endosulfan sulfate, were also detected at most of these sites. Gamma chlordane (1.3 - 3.5 ng/g) was detected at 5 sites and DDD (2.5 - 6.8 ng/g) was detected in sediment from 6 sites. The concentration of 76 ng endosulfan I /ℓ in water measured at site C111E-1, above structure S-178, exceeds the freshwater chronic criteria value (56 ng/ℓ) and is approximately 1/3 of the freshwater acute criteria value of 220 ng/ℓ. Methyl mercury concentrations in water from Florida Bay tended to be higher (55 - 480 pg/ℓ) than those measured at the canal sites (<50 - 107 pg/ℓ) during the March sampling. Total mercury concentrations in sediments ranged from 10 - 99 ng/g for all sites.

464 Hazard Assessment of Mosquito Control Insecticides in South Florida Coastal Ecosystems. R. H. Pierce and M. S. Henry, Mote Marine Laboratory, Sarasota, FL and G.M. Rand, Florida International University, Miami, FL. The distribution and persistence of mosquito control pesticides was determined following aerial applications directly into or adjacent to coastal ecosystems. The larvicide, temephos, was monitored following applications directly into saltmarsh communities. The adulticides, fenthion, permethrin and naled, were monitored as drift into coastal ecosystems following aerial and ground application over residential areas adjacent to the non-target study sites. All studies were performed during actual mosquito control applications under natural field conditions. In addition, a hazard assessment was also conducted including malathion and resmethrin using the U.S. EPA framework. Results showed that an ecological hazard resulted for several non-target organisms. When exposure and fate characteristics are considered, certain insecticides which have high acute toxicity exhibit a low hazard to aquatic organisms. Changes in the application techniques used by the mosquito control districts were found to successfully reduce the ecological hazard, while maintaining adequate mosquito control.

465 Ecotoxicological Risk Assessment Models of Urban and Agricultural Nonpoint Source Pesticide Runoff Impacts on Living Marine Resources of Florida Bay. G. Scott, M. Fulton, J. Kucklick, E. Strozier, P. Pennington, M.DeLorenzo, P. Key, J. Daugomah, K. Chung, and E. Wirth, NOAA, NOS, Charleston Laboratory; J. MaCauley, J. and L. Goodman, EPA, GCEL; G. Thayer and M. LaCroix, NMFS, SEFSC Beaufort Laboratory; and G. Chandler, School of Public Health, USC. Field and laboratory research was conducted from 1993-98, to assess the potential ecotoxicological effects of urban and agricultural pesticides via nonpoint source (NPS) runoff on the living marine resources of Florida Bay. Field studies were focused on quantifying the levels and distribution of urban and agricultural pesticides in water, sediment and biota (oysters, fish and SPMDs.) In addition the potential acute and chronic toxicity of agricultural NPS runoff on *Gambusia affinis, Palaemonetes pugio and Crassostrea virginica* was evaluated, so that a potential risk assessment characterization could be made. Laboratory toxicity testing was focused on *P. pugio, Amphiascus tenuirernis* and *Mercenaria mercenaria* in marine habitats and *Daphnia magna, Selanastrum* and microbial loop communities in freshwater habitats. Results indicated significant runoff of endosulfan and other pesticides into the C111 canal and surface waters of Florida Bay. Surface waters contained mixtures of several pesticides, many of which were potential endocrine disrupting chemicals. These compounds were measured in fish, oysters and SPMDs with trophic transfers in biota that were predictable by Fugacity Modeling. Sediment toxicity tests with copepods and *in situ* oyster bioassays found significant alterations in reproduction. Both decreased (herbicides) and enhanced (nutrient enrichment)growth was observed in *Selanastrum* and decreased survival in *Daphnia magna*. These results indicate that pesticide and nutrient inputs into Florida Bay may have a significant impact on living marine resources of Florida Bay.

466 Fate and Effects of the Insecticide/Miticide, Pyridaben using a Freshwater Outdoor Microcosm System. Rand, G.M.*, Florida International University, Miami, FL; Clark, J. R. and Holmes, C. M., BASF Corporation, Research Triangle Park, NC. An ANOVA design was used with four treatments in which Pyridaben, was directly applied to water of 18 tanks at three different concentrations (0.34, 3.4, 34.0 μg/L) and only water was applied to six control tanks. Pyridaben was applied two times in the morning below the water surface with a 30-day interval in between. Chemical and biological monitoring were conducted during a 11-month baseline phase followed by a total of seven weeks for treatment/post-treatment phases.The half-life in water for the low, mid, and high treatment concentrations for both applications ranged from 11.8-28.5 hours which supports laboratory photolysis studies. Following both applications at 3.4μg/L it was not detectable in sediment within 24 hours while at 34.0μg/L, the half-life in sediment was 9.8 days. Copepoda abundance was significantly reduced at 34.0μg/L but recovery occurred within eight weeks after application. Copepoda nauplii abundance was significantly reduced at 3.4 and 34.0 μg/L; effects were more severe at 34.0μg/L and recovery was more rapid at 3.4μg/L. Rotifer abundance was reduced at 34.0μg/L and recovery occurred within eight weeks for all groups except *Polyartha* and *Macrochaetus*. Phytoplankton, periphyton, and macro invertebrates were not significantly affected. Pyridaben was acutely toxic at 34.0μg/L to bluegills but not toxic at the 96-hour LC50 (3.4μg/L). Based on the results of laboratory and microcosm data, Pyridaben will not present a hazard to aquatic organisms when applied at typical agricultural rates.

467 A Comprehensive Assessment of Agriculturally-Influenced Florida Bay Sediments Using a Benthic Copepod Reproduction Model. Chandler, G.T.*, Klosterhaus, S.L., University of South Carolina, Columbia, SC, Scott, G.I. and Fulton, M.H., NOAA/NOS, Charleston, SC. Benthic copepod partial life-cycle bioassays (*Amphiascus tenuiremis*) were concurrently performed on pristine North Inlet, SC, clean sediments, Florida Bay clean control sediments, and 10 other slightly to moderately pesticide-contaminated sediments from in and around the Florida Bay estuary. A number of test endpoints were compared: Male:female survival, no. of females gravid at the end of the tests (i.e. EOT), no. of nauplii and copepodites produced alive at EOT, reproductive potential at EOT, and realized reproductive output at EOT. Only one site produced significant adult mortality (30%:Joe Bay), but several sites produced significantly depressed reproductive potential and output. Reproductive effects were most strongly focused on offspring production & survival, rather than clutch size. Naupliar production was significantly reduced 35 to 85% at 5 sites, copepodite production was significantly reduced at all sites but the control, and was reduced > 93% at 6 of 10 sites. Significantly depressed mean clutch sizes were seen for Manatee Bay (22-29%). At the end of each bioassay, potential reproduction was conservatively assessed for each replicate sediment culture as (total eggs + total nauplii + total copepodites)/(no. surviving females). Potential reproduction was significantly depressed in Joe Bay, Manatee Bay and Little Madeira Bay, suggesting that continued culture in these sediments would have led to significantly depressed population growth through time. Actual realized production (total nauplii + total copepodites)/(surviving females) after 14-d culture showed depressions of 40-100 % of controls in Joe Bay, the C-111 canal, Manatee Bay and Little Madeira Bay. Chemical analyses revealed the presence of chlorpyrifos, endosulfan and other pesticides at various concentrations.

468 Factors Affecting Bioavailability of Methyl Mercury in Florida Bay. Lores, E.M.*, J. Macauley & L.R. Goodman, U.S. EPA, Gulf Breeze, FL; R.G. Smith and D.M. Wells, Skidaway Institute of Oceanography, Savannah, GA. A field sampling study was designed to investigate the factors affecting concentrations and availability of Methyl Mercury (MeHg) in Florida Bay. Sampling effort was concentrated along the mangrove fringe from the mouth of Taylor River to Barnes Sound but included a transect across Florida Bay from Trout Creek to Key Largo. A preliminary sampling in June, 1995, found MeHg in filtered water samples ranged from <10 to 2,300 pg/L while total Hg in fish tissue ranged from 0.034 to 41 ug/g. In 1997, filtered and ultrafiltered samples were collected during dry and wet seasons (March and September). MeHg and DOC were measured in filtered (0.45 um) and ultrafiltered (1000 Dalton) samples collected on site. During the dry season, concentrations of MeHg ranged from <50 pg/L to 575 pg/L in filtered samples and, on average, less than 30% passed through the ultrafiltration membrane. DOC concentrations ranged from 5.7 to 11 mg/L. Wet season MeHg concentrations ranged from 61 to 395 pg/L in filtered samples with approximately 50 % passing through the ultrafiltration membrane. At Taylor River, over 95% was retained by the ultrafiltration membrane during both wet and dry seasons.

469 Commonalities in Mercury Concentrations Among Largemouth Bass Populations from the Southeastern U.S. Engel, D.W.*; Evans, D.W., NOAA/NMFS/ SEFSC, Beaufort Laboratory, Beaufort, N.C. Elevated mercury concentrations occur among freshwater fish in the southeast, including the largemouth bass, *Micopterus salmoides*. Concerns about mercury in fish tissues are based on the human fish consumption where action limits for methyl mercury concentrations in edible tissue are set >0.5 or >1.0 ppm, depending on the state. In most areas, where mercury is a concern,affected fish tend to be remote from urban and industrial sources and are not associated with known point sources of mercury. Aerial deposition is implicated as the most probable source. In addition to a shared diffuse source, high mercury concentrations are associated with common water quality characteristics. A strong negative correlation exists between water chemistry (pH and alkalinity), productivity, and mercury concentrations in largemouth bass across all states. Data collated from the different states indicates that elevated methyl mercury concentrations in bass also are related to their food web relationships. The identified factors associated with bioaccumulation of methyl mercury by freshwater fish in remote locations appear to be aerial deposition of mercury, water chemistry, similar life history strategies, and food web relationships which are common to all coastal plain environments in the southeast from North Carolina to Texas.

470 The Role of Coral Diseases and Anthropogenic Stressors on Tropical Marine Coral Reefs. Santavy, D.L.*, Campbell, J.G. US EPA, Gulf Breeze, FL. Stony (scleractinian) and soft (octocorals) corals throughout the Western Atlantic have been affected by several fatal diseases in the last two decades. In many locations the communities have not recovered from these diseases and the ecosystem has permanently changed. Several hypotheses have attempted to explain these the causes of the destructive events. These hypotheses have ranged from deleterious effects of anthropogenic inputs to epizootics to ariel deposition of Saharan particulates. In the Florida Keys, the coral reef ecosystem has experienced changes in water quality from extreme coastal development resulting in landscape modifications. Increased levels of nutrients, metals, pesticides, herbicides, and particulates are entering into the marine environment in this region. It is clear that corals, a rare and protected resource, are exposed to increasing physical, chemical, biological and climatic assaults. Concurrently, increases in coral tissue loss, lesions, neoplasms, and mortality have been observed throughout this same region. Studies in the Lower Florida Keys, the New Grounds, and the Dry Tortugas aim to establish baseline information on the occurrence of these diseases and relate them to potential stressors. Due to restrictions on coral collection in national sanctuaries, sponge species are being used as surrogates to determine potential contaminant bioaccumulations in corals. Previous studies have shown corals to be more sensitive to allochthonous substances than organisms from temperate estuarine and marine systems. Therefore it is becoming apparent that a lack of knowledge on baseline data, indicator and biomarkers species, bioavailability and bioaccumulation for tropical marine environments are hampering our ability to determine the conditional status of the ecosystem and measure its recovery after major assaults.

471 Laboratory and Field Analyses of the Potential Toxicity of Polydimethylsiloxane (PDMS) to Benthic Macroinvertebrates. Henry K.S.*[1], Nocentini, A.[2], Wieland, W.[3], Rossi, D.[2], Beltrami, M.[2], Baudo, R.[2], and Giesy, J.P[1]. [1]Michigan State University, East Lansing, MI, [2]Italian Institute of Hydrobiology, Verbania Pallanza, Italy. [3]Agricultural University Wageningen, The Netherlands. Polydimethylsiloxane (PDMS) is widely used in a number of industrial processes and consumer products that result in down-the-drain disposal. Negligible volatility and water solubility, and a slow degradation rate, suggest that PDMS may accumulate in sediments. Measurable concentrations of PDMS are observed in some surface sediments near outfalls of wastewater treatment facilities. Previous laboratory exposure studies found no acute or chronic toxicity of PDMS to a variety of benthic invertebrates. In the present study, the amphipod *Hyallela azteca* and the midge *Chironomus tentans* were exposed to PDMS-treated sediments of varying organic matter content in the laboratory during short- and long-term exposures. Endpoints for short-term tests included survival and growth, while chronic assays considered survival, growth, reproduction, and emergence. Potential effects of PDMS on benthic communities were investigated *in situ* in plastic trays containing natural sediments that had been defaunated and spiked with PDMS to concentrations of 1000 ppm (nominal d.w.). Trays were placed at 10 m depth in Lago Orta, Italy, a lake which has been historically contaminated with ammonia and heavy metals. Sediment-containing trays were also placed at 14 m in a Michigan lake which has been developed along its shoreline, but which does not receive direct contamination. At least one tray at each PDMS concentration was collected at seasonal intervals from both lakes and invertebrates sieved and identified to genus or species, when possible. Field studies and acute and chronic laboratory exposures indicated that environmentally relevant concentrations of PDMS did not significantly impact community structure, survival, or growth.

472 St. Clair River Sediments Part 2 - Fathead Minnow Toxicity. Kierstead, T., Nova Chemicals, Sarnia, Ontario; Moran, T.*, Walker, K., Pollutech EnviroQuatics Ltd, Sarnia, Ontario; Munro, T.S., Lambton Industrial Society, Sarnia, Ontario. An integrated assessment of sediments in the northern reaches of the St. Clair River included a 21 day acute toxicity test using fathead minnows (*Pimephales promelas*). Results demonstrated acute toxicity to fathead minnows at eight of 28 study locations and 1 of 3 reference locations. Three other test species - *Hexagenia spp., Chironomus riparius,* and *Tubifex tubifex* - showed no effect, or sublethal/chronic effects only, at the same locations. To test speculation that lethality to fathead minnows was due mainly to the laboratory static toxicity test method, two additional test methods were developed. Exposures were repeated using the standard fathead minnow 21 day static test; a laboratory 21 day flow-through test using river water as overburden replacement water; and, a 21 day in-situ test in which fathead minnows were caged on the river bottom in direct contact with the sediments at the original locations where acute fathead minnow toxicity occurred. Overburden water in the static test was analysed at the beginning and end of the test. Lethality was significant in the static tests, but did not differ significantly from controls in either the flow-through or in-situ tests. Analysis of the static overburden water suggested that lethality was linked to ammonia, possibly iron and possibly neutral extractable chlorinated hydrocarbons accumulating in the overburden water depending on location. The study concluded that these contaminants remain below measurable effect levels in the river. Results of the in-situ tests, if applied to the interpretation of the fathead minnow results, has the ability to profoundly affect earlier integrated assessment conclusions for the St. Clair River.

473 Regional Patterns and Scope of Toxicity in Estuarine Sediments. Long, E. R.* National Oceanic and Atmospheric Administration, Seattle, WA. Hameedi, M. J., National Oceanic and Atmospheric Administration, Silver Spring, MD. Laboratory tests of the toxicity of estuarine sediments were performed from 1990 to the present in 25 estuaries and bays along the Atlantic, Pacific, and Gulf of Mexico coastlines as a component of the National Status and Trends Program. These tests were performed to determine the severity, spatial gradients, and surficial extent of toxicity as a measure of adverse biological effects attributable to the presence of toxicants. Tests of solid-phase sediments were performed with amphipods, tests of pore waters were performed with sea urchin gametes and embryos, and tests of solvent extracts were performed with both Microtox and cytochrome P-450 RGS assays. Data from these surveys showed a wide range in toxicity among the different survey areas and among tests. This paper will summarize results obtained thus far by NOAA and compare them to results obtained in EMAP studies conducted within estuarine provinces. Also, the paper will illustrate spatial gradients in toxicity within selected areas and rank the spatial extent of toxicity among areas.

474 Chemical and Toxicological Characterization of Rybinsk Reservoir Sediments. Cleveland, L.*, USGS, Environmental and Contaminants Research Center, Columbia, MO, USA, Tomilina, I. I., Flerov, B. A., Gapeeva, M.V. and Bakanov, A. I., RAS, Institute for Biology of Inland Waters, Borok Russia. Ten surface water and sediment samples were collected from sites extending from the northern shore of the Rybinsk Reservoir to south of the Darwin Reserve. Toxicity of the surface water samples was measured in forty-eight hour tests with *Ceriodaphnia dubia*. The whole sediment samples were physically and chemically characterized, examined for the presence of benthic organisms, and tested using 7-day elutriate tests with *Ceriodaphnia dubia* and 14-day whole-sediment tests with *Hyalella azteca*. Across the sampling sites, total PAHs ranged from 43 to 0.27 μg/g, dry weight; PCBs ranged from 11 to 0.026 μg/g, dry weight; and total metals ranged from 2470 to 706 μg/g, wet weight. Adverse effects in the toxicity tests were more severe for the northern sites in the reservoir and the effects begin to diminish at sites about 55 km south of the city of Cherepovets. Diversity and biotic indices determined for each site were integrated to derive a Combined Index of Community Status (CICS). The CICS could be useful in distinguishing effects on benthic communities due to pollution and in ranking the status of benthic communities over large areas. This study implicates the highly industrialized city of Cherepovets on the northern shore of the Rybinsk Reservoir as a significant source of pollutants that contaminate the reservoir up to 55.

475 **The Use of Chronic Sublethal Bioassays to Determine the Spatial Distribution of Toxicity in the Bush River, MD.** Emery, Jr., V., Moore, D. USAE, Waterways Experiment Station, Vicksburg, MS, Wright, R.B. AScI Corporation, Vicksburg, MS. Chronic sublethal sediment bioassays were conducted to determine the spatial distribution of toxicity in the Bush River and its tributaries using the estuarine amphipod *Leptocheirus plumulosus*. Two hundred twenty eight sediment samples were collected covering an approximate 175 square mile area from 38 discreet zones in the Bush River bordering the Aberdeen Proving Grounds-Edgewood Area. Toxicity was determined following 28-d amphipod exposures to field replicated sediments (6 samples/area) in a one-way block design. The endpoints evaluated were survival, growth, and reproduction. Survival ranged from 55 to 92%, growth (mg individual dry weight/day) from 0.42 to 0.89, length (mm) from 5.8 to 7.8, and reproduction (number of neonates/surviving adult) from 0.34 to 2.6 . Linear contrast analyses were used to detect toxicity gradients between the tested zones. Covariate analyses were used to evaluate the potential correlations between sediment grain size, TOC, and toxicity. In addition, measured concentrations of sediment associated contaminants were compared to published ER-Ls, ER-Ms, and EPA SQALs to evaluate the utility of numerical sediment criteria in predicting observed sediment toxicity.

476 **Conformance of Sediment Quality Guidelines with Measured Toxicity.** O'Connor, T.P.* NOAA, Silver Spring, MD, Hyland, J. L. NOAA, Charleston, SC, Paul, J. F. US EPA, Narragansett, RI, Summers, J. K., US EPA, Gulf Breeze, FL. Sediment surveys conducted by NOAA and EPA provide a data set of 1700 samples with which to test proposed relationships between sediment chemistry and toxicity. If toxicity is defined as less than 80% survival of amphipods during 10-day exposures to whole sediment, 13% of the samples were toxic. Estimates based on equilibrium partitioning predicted toxicity in only 12 samples and half of those were not toxic. About 13% of the samples had at least one chemical concentration above a bulk chemical-based ERM guideline, but only a third were actually toxic. Guidelines that predict a lack of toxicity faired better. Almost 31% of all samples did not exceed the bulk chemical-based ERL and 95% of those were not toxic. Of the 1375 samples with AVS measurements, 74% had SEM/AVS (simultaneously extracted metal/acid-volatile sulfide) ratios less than 1.0 implying no toxicity from trace metals. Twelve percent were toxic but not necessarily because of trace metals. It was found that this guideline is more sensitive to AVS than to metal contamination. The various guidelines can serve to trigger further analysis, but none should be used, alone, to identify toxic sediments or to make decisions on sediment management.

477 **An Additive Chemical Mixture Evaluation of Sediment Toxicity Test Results for Three Amphipod Species.** Fuchsman, P.C.*; Barber, T.R.; Duda, D.J., McLaren/Hart, Inc., Cleveland, OH; Wenning, R.J., McLaren/Hart, Inc., Alameda, CA. Toxicity test results for three amphipod species exposed to sediment from a Gulf Coast estuary were evaluated using a site-specific, additive chemical index. No-effect concentrations for individual chemicals of interest were developed from site-specific toxicity test results (hexachlorobutadiene and mercury) and studies reported in the scientific literature (chlorinated benzenes, polycyclic aromatic hydrocarbons, copper, lead, and zinc). Evaluation of sediment chemical concentrations on an individual-chemical basis explained most toxicity observed in the data set used initially to evaluate hexachlorobutadiene and mercury (*Hyalella azteca* and *Leptocheirus plumulosus* tests) and in an independent site-specific set of sediment toxicity test results (*Ampelisca abdita* tests). An additive model was developed using the no-effect concentrations for individual chemicals of interest to account for possible effects due to chemical mixtures. The additive index showed a logistic concentration-response relationship with amphipod survival for both data sets. The index values below which toxicity was never observed (2) and above which toxicity was always observed (8.5) were identified as site-specific no-effect and effect concentrations for chemical mixtures. The likelihood of sediment toxicity corresponding to intermediate index values is uncertain and depends in part on site-specific conditions affecting the bioavailability of metals.

478 **A Comparison of Sediment Bioassay Protocols Involving Contaminant Mixtures.** Van Dolah, R.F., Maier, P.P., Jones, J.D., Ringwood, A.H., Keppler, C.J., Conners, D.E., SCDNR-Marine Resources Division, Charleston, SC; Fulton, M.H., Scott, G.I., Chung, K., National Ocean Service, Charleston, SC; G.T. Chandler, University of South Carolina, Columbia, SC; Lee, R.F., Skidaway Institute of Oceanography, Savannah, GA; Snell, T.W., Cecchine, G.A., Georgia Institute of Technology, Atlanta., GA. Sediments from four tidal creeks (3 polluted, 1 reference) were tested to evaluate the relative sensitivity of nine bioassay protocols. Five protocols exposed organisms (*Palaemonetes pugio, Ampelisca verrilli, Amphiascus tenuremis, Mercenaria mercenaria*) to sediments for 7-14 days, with mortality or growth as the toxicity measure. Four protocols exposed organisms (*Brachionus plicatilus, Callinectes sapidus, Crassostrea virginica*) to sediment elutriate or porewater for 2-48 hrs, with population growth or fertilization/egg development as the toxicity measure. The polluted creeks contained mixtures of both metal and organic contaminants that exceeded bioeffects guidelines. Mean ER-M quotients at those sites ranged from 0.67 to 0.13. Bioassay results suggested that the *C. sapidus* and *M. mercenaria* assays were most sensitive at some of the sites and the *P. pugio* and *B. plicatilus* assays appeared to be the least sensitive. None of the assay protocols showed consistently greater sensitivity at sites where bioeffects were anticipated.

479 **Limitations and Uncertainties of Sediment Effects Concentrations (SECs) in the Evaluation of Sediment Toxicity and Chemistry.** Anid, P.J.*, HydroQual, Inc. Mahwah, NJ; Connolly, J.P., Quantitative Environmental Analysis, Inc. Montvale, NJ. Limitations and uncertainties of SECs values were evaluated using two major cases of contamination in the Hudson-Raritan Estuary (HRE) and Los Angeles Harbor (LAH). These screening values, determined by matching biological with contaminant concentration data are not indicative of cause-effect relationships between a single contaminant and toxicity, and often lead to serious over- or underestimation of toxicity. The analysis of HRE and LAH data show that SECs were not good predictors of toxicity, in particular, at low or medium contamination. With the exception of hot spots areas, most environmental concentrations of PCBs and PAHs seem to fall within this low to intermediate range, and covariance among contaminants (i.e.; mostly PCBs, PAHs, pesticides and metals) seems to weaken the predictive ability of the screening values. The occurrence of significant overlapping in the range of concentrations exerting both toxic and non-toxic effects and the consequent high probability of type I and type II errors seem almost unavoidable. As a result, none of the screening values provide high reliability for the assessment of toxicity of sediment-associated contaminants. Because narcosis values are driven by the chemical and physical properties of the contaminant rather than by the presence of other chemicals, our analysis indicates that narcosis might be a better approach for evaluating toxicity of individual toxicants. Based on circumstantial evidence, when PCBs or PAHs are the dominant

contaminants, toxicity seems to better match narcosis level than ERM values, especially in the case of PCBs. Although it does not account for possible synergistic, antagonistic or additive effects, the narcosis approach seems promising for the development of SQCs that represent potential toxicity of specific compounds.

480 The Use of Site Specific Sediment Toxicity Testing to Establish Sediment Action Criteria. Walter, B.R., Woodard & Curran, Portland ME; Ostrye, D.P. ,Woodard & Curran, Portland, ME; Hathaway, E.M., U.S.EPA, Boston, MA; Truchon, S.P., Exponent, Waltham, MA; Williams, J.W., AquaTech, Burlington, VT. Metal contaminated groundwater at the Saco Municipal Landfill Superfund Site, in Saco, Maine was found to be discharging to a local stream (Sandy Brook). Studies found that naturally occurring metals (primarily arsenic, iron and manganese), were being leached from the underlying bedrock due to reducing conditions induced by the leachate discharge from the landfill. The leached metals discharged to Sandy Brook where changes in water chemistry resulted in the precipitation of the leached metals. Sediment sampling in the Brook and adjacent wetlands detected concentrations as high as 2,250 mg/kg arsenic - several hundreds times the 8.2 mg/kg ER-L for arsenic. A Sediment Removal Action was completed in wetlands immediately adjacent to the brook using a conservative interim cleanup concentration of 19 mg/kg arsenic. Approximately 200 yd^3 of sediment were removed. However, it became evident that 19 mg/kg arsenic for the stream sediments was not attainable without significant disruptions to the existing aquatic ecosystem. Aside from a visible iron floc, the Brook did not appear to be adversely impacted, suggesting that site-specific factors may be limiting the potential toxic effects of arsenic. Acute and chronic laboratory sediment toxicity tests were conducted using the amphipod *Hyalella azteca* in conjunction with a baseline ecological risk assessment to determine environmentally acceptable endpoint concentrations. Preliminary acute toxicity data suggest that acceptable endpoint arsenic concentrations of 100 mg/kg or higher may be appropriate. Final results of the acute and chronic tests, available in mid-July will be discussed along with resulting site restoration strategy.

481 Establishing Causal Relationships Between Biomarkers and Higher-level Responses along a Contaminant-loading Gradient in Polluted Streams. Adams, S.M., Greeley, M.S., and Ryon, M.G., Oak Ridge National Laboratory, Oak Ridge, TN. Effects of environmental stressors such as contaminants on the health of fish populations usually involve a series of biological responses ranging from the biomolecular/biochemical to the population and community levels. Biomarkers of environmental stress at lower levels of biological organization such as MFO detoxification enzymes and DNA integrity provide direct evidence of contaminant exposure while intermediate-level responses such as histopathological, bioenergetic, and reproductive changes can be predictive of effects at the individual, population, and community levels. Responses at the lower levels of biological organization have the primary advantage of being relatively sensitive (short-term response) to stressors thus serving as early warning indicators of potential environmental degradation. Conversely, responses at higher levels of organization are relatively insensitive (long-term response) to stressors but have higher ecological relevance and are therefore more directly applicable to the ecological risk assessment (ERA) process. Biomarkers, however, cannot be considered bioindicators of ecosystem stress or effectively applied in the ERA process unless they are causally linked or related to ecologically relevant endpoints such as population or community level indicators. To determine causal relationships between lower (i.e. biomarkers) and higher level effects, responses in fish populations at several levels of biological organization were measured along a contaminant gradient in two polluted streams. The downstream response patterns for several biomarkers along this contaminant gradient were similar to higher-level response patterns indicating that these biomarkers could be potentially useful in the ERA process because of their sensitivity and relationships to ecologically relevant endpoints.

482 Relationship Between Genotoxicity, Mutagenicity, and Community Structure in a Contaminated Stream. Theodorakis, C.W.*, Swartz, C., Rogers, J. and Bickham, J.W. Texas A&M University, College Station, TX; Adams, S.M., Oak Ridge National Laboratory. This study was conducted in to determine (1) genotoxic and cellular biomarker responses of native fish populations exposed to industrial effluent, (2) mutagenicity of the associated sediments and (3) relationships between biomarkers, mutagenicity, contaminant concentrations, and community-level responses. Redbreast sunfish (*Lepomis auritis*) and stoneroller minnows (*Campostoma anomalum*) were collected from a reference stream and East Fork Poplar Creek (EFPC), a first-order stream which originates on the grounds of the Department of Energy Y-12 Plant at Oak Ridge, TN, and is contaminated with mercury, PCBs, and numerous other compounds. Previous studies have shown that sediment contaminant concentrations, as well as physiological biomarker responses of the local fish populations, are highest at the headwaters of EFPC and decrease with increasing distance from the DOE facility as contaminant loading decreases. Biomarker responses were determined by flow cytometry, and included variation in cellular DNA content (an index of genotoxicity) and perturbations in the cell cycle of blood and kidney tissues. Mutagenicity was determined by the Ames assay using organic solvent extracts of sediment surface samples. Community level responses included the Index of Biotic Integrity and community diversity. Biomarker responses and mutagenicity were found to be highest at the headwaters of EFPC, and decreased with increasing distance from the effluent. In general, biomarker responses appeared to be correlated with mutagenicity of the sediment, and both of these related to fish community disturbance and level of stream contamination. Because responses at several levels of biological organization show similar patterns of downstream effects, this suggests that there may be a causal relationship between contamination and biological effects.

483 Ordinating Sensitivity of Bioindicators of Persistent Contaminant-Induced Stress: Multitier Responses of Chronic Applications of Creosote. Ciborowski, J.J.H.*, University of Windsor, Windsor, ON; Solomon, K.R., Harris, M.L., Bestari, J.; Day, K.E., Environment Canada, Burlington, ON; Marwood, C., University of Waterloo, Waterloo, ON. Environmental perturbations can be chronic or acute and may produce different biological effects. A "pulse" is a single, strong, event that produces an acute shock to the system, analogous to a toxicant spill. Ecotoxicological studies typically mimic pulse events. A "press" study is characterised by chronic application of (frequently low-level) stress. We studied responses over 95 days across levels of organization (subcellular, cellular, organismal, population, community) in plankton, benthos, macrophytes and fishes reared in 10,000-L microcosms that received either multiple low-concentration or single high-concentration injections of creosote, a complex mixture of hydrocarbons and PAHs. Previously, we had found common toxicity thresholds (EC_{50}) of 1-5 mL/L in response to single (pulsed) applications of creosote (*e.g.*, chironomid larvae - deformities 1.0 mL/L, abundance 1.2 mL/L, genus richness 5.3 mL/L; *Myriophyllum* - fluorescence 3 mL/L, growth 3 mL/L; fathead minnows; follicular development insensitive, LC_{50} 1 mL/L). Toxic responses to the pulse were typically manifested too quickly for a 'best' (most sensitive) bioindicator to be identified. Gradual accumulation of contaminant in press exposures should better separate time to response of more sensitive (early responding) from less sensitive (later responding) bioindicators. We anticipated that subcellular or cellular processes would be more sensitive sublethal indicators of contaminant stress, but this was not always the case. Chronic exposure induced toxic responses at concentrations consistently lower than occurred in response to pulse exposure. There was little evidence of acclimation to chronic stress relative to responses seen in pulsed contaminant applications.

484 Assessing the Ecological Characteristics of Wetlands Receiving Oil Sands Effluent. Bendell-Young, L. I. Bennett, K. E., Crowe, A., Farrell, A. P., Kennedy, C.J., Kermode, A. R., Moore, M.M., Plant, A. L., Wood, A. Dept. of Biological Sci. Simon Fraser University, B.C., Canada. The primary objectives of this study were to evaluate the ecological characteristics of wetland ecosystems that had developed in response to oil sands effluent relative to reference wetland ecosystems and from such an evaluation, to assess if these wetlands were viable systems capable of integrating into the northern Canadian landscape. A secondary objective was to evaluate the use of several ecologically relevant endpoints as indicators of an ecosystem response to a known anthropogenic stress, in this case, wetlands receiving oil sands effluent. To achieve this, a suite of endpoints were compared between effluent-impacted wetlands and non-impacted reference wetlands. Endpoints for comparison included: 1/ benthic macroinvertebrate community structure, 2/ chironomid density and biomass, 3/ the incidence of chironomid mentum deformities, 4/ the mutagenetic potential of sediment-dwelling chironomids, 5/ growth and photosynthetic rate for the aquatic plant *Typha latifolia*, (cattail) and 6/ fish acute lethality and stress response as measured by changes in blood chemistry (%haematocrit (%Hct.), %leucocrit (%Lct.), and differential white blood cell count). Wetlands receiving oil sands effluent supported a low diversity benthic community, dominated primarily by the Chironomidae and cattail. There was no evidence of mentum deformities or mutageneticy in chironomids sampled from the oil impacted wetlands. Cattails grown in oil sands effluent and sediment demonstrated increased photosynthetic rates; however, these increased rates did not translate into increased plant growth. In contrast to the benthic community and the cattail, indigenous fish were unable to survive in wetlands containing oil sands effluent. Fish displayed altered blood chemistry (elevated %Hct., depressed %Lct.) and ultimately death when held beyond 14 days in the oil-impacted wetlands. Of the various ecological endpoints measured, the macroinvertebrate community and changes in fish blood chemistry were the most sensitive indicators of an anthropogenic stress, demonstrating distinct differences in response between impacted and reference wetlands. To ensure that these wetlands can safely integrate into the northern Canadian landscape, future studies need to focus on their impacts at higher trophic levels indigenous to the wetland.

485 Toxicity of Pesticide Mixtures in Aquatic Environments: Assessing Effects Across Several Levels of Biological Organization. Sibley, P.K.*, Chappel, M., and Solomon, K.R., Centre for Toxicology, University of Guelph, Guelph, Ont.; George, T. and Liber, K., Toxicology Centre, University of Saskatchewan, Saskatoon, Sask.; Ciborowski, J., Dept. of Biology, University of Windsor, Windsor, Ont. In 1997, we initiated a comprehensive study to evaluate the utility of using a probabilistic risk assessment approach to examine the toxicity of chemical mixtures. As a first step in the evaluation, we assessed the toxicity of binary and tertiary mixtures of organophosphorus insecticides (chlorpyrifos, azinphos-methyl, diazinon) in outdoor microcosms using a regression design based on a concentration gradient determined from analysis of the 90th centile exposure distribution data for each compound. Impact was assessed by measuring response variables at several levels of biological organization, including acetylcholinesterase (AchE) inhibition in fish and invertebrates (biochemical), mortality of fathead minnows (individual), and changes in phytoplankton, zooplankton, and benthic communities (population). AchE was significantly inhibited within 2-4 d of exposure to the lowest concentration of OP mixture in both fish and the cladoceran *Bosmina*. Temporal changes in AchE inhibition were strongly correlated with changes in zooplankton population abundance, indicating that the former could be used to predict the latter. An increase in phytoplankton and micro-zooplankton (Rotifera) abundance indicated a release of these groups from competitive interactions with the zooplankton due to selective toxicity of the OP's to zooplankton. AchE inhibition was also strongly correlated with fish mortality, although at higher concentrations than with invertebrates. The results of this study indicate that measurements conducted at specific levels of biological organization (e.g., AchE) can be used to adequately predict effects at higher levels of organization (e.g., individual, population), and emphasize the importance of incorporating multiple measures of effect, across several levels of biological organization, to undertake effective risk assessments.

486 Does an Association Between 4-nonylphenol Use and Subsequent Declines in Catch of Atlantic Salmon (*Salmo Salar*) Represent a Case of Endocrine Disruption? Fairchild, W.L.*, Taylor, E.O., Arsenault, J.T., Gulf Fisheries Centre, Moncton, NB, Canada; Brown, S.B., National Water Research Institute, Burlington, ON, Canada. Historical aerial applications of the insecticide formulation Matacil® 1.8D provide a unique opportunity to look for potential population level effects of the endocrine disrupting compound 4-nonylphenol (4-NP) on Atlantic salmon (*Salmo salar*). Matacil® 1.8D contains the carbamate insecticide aminocarb with 4-NP as a diluent. Between 1975 and 1985 it was applied to large forested areas of Atlantic Canada to control damage from spruce budworm (*Choristoneura fumiferana*). After spraying, aminocarb concentrations in water samples were well below lethal thresholds for salmon. However, estimated 4-NP concentrations fell within a range where estrogenic effects might be anticipated. The spraying coincided with final stages of smolt development in Atlantic salmon. Smoltification is a critical time for home stream imprinting, development of hypo-osmoregulatory abilities, and occurs in association with changes in several hormonal systems. Salmon catch data were evaluated considering effects on survival at the smolt stage. There was a significant negative relationship between the returns of large Atlantic salmon and the proportion of individual tributaries sprayed within the Restigouche River drainage basin in 1977. There was also a broader event of unusually heavy salmon smolt mortality in 1977. This smolt mortality event contains a significant relationship indicating that where Matacil® 1.8D spraying occurred, the smolt mortality increased. For 14 rivers exposed to spraying, 53 % of the lowest salmon catches between 1973 and 1990 coincided with times when Matacil® 1.8D had been sprayed. We hypothesize that the 4-NP in the Matacil® 1.8D formulation is the causal agent. Concentrations of 4-NP described here are within ranges encountered in the environment today.

487 Evidence of Metal Effects from Long-term Biomonitoring at a Site in San Francisco Bay. Luoma S., Hornberger M, Cain D, Parcheso F., Thompson, J. U.S. Geological Survey, Menlo Park, CA. Metal pollutants could affect ecosystems by eliminating metal-sensitive species, eliciting compensation or stress in survivor species, and allowing proliferation of tolerant species. Of these effects, changes in survivor populations may provide the least ambiguous evidence of metal influences. This paper reconstructs a pattern of benthic effects and recovery from Cu and Ag contamination using a 22 year data record from a site in San Francisco Bay (SFB). Monthly analysis of metals between 1975 and 1997 identified long-term trends in exposure to Cu and Ag in the bivalve, *Macoma balthica*, with substantial seasonal fluctuations in bioaccumulated concentrations. Annual maximum Cu in clams declined from >500μg/g in 1980 to 70μg/g in 1997. Annual maximum Ag concentrations were >200μg/g in 1978 to 9μg/g in 1997. Early studies indicated biochemical stress during the period of high metal exposure. Reproductively mature gonadal cells were rarely present in sections from archived animals collected when contamination was greatest. The most severe effects on reproduction occurred in the Fall when metal concentrations were increasing. Simple trends in condition index were not evident, but detrended analysis of seasonality showed that weight gain in the Fall reproductive season appeared after Cu and Ag concentrations declined. Skewed gender ratios were observed in the early 1980's, but not in later collections. Adult clam populations were Cu- and Ag-tolerant in the early 1980's relative to uncontaminated populations, facilitating the presence of the species. Immigration probably also facilitated survival. Metal contamination unequivocally affected biochemical, physiological and population processes in *M. balthica* through the 1980's; as evidenced by recovery from those effects as metal exposure declined.

488 Characterizing the Aquatic Health in the Boulder River Watershed, Montana by Integrating the Exposure of Food Web Organisms with Fish Physiology and the Abundance of Wild Fish. Farag, A.M.*, USGS - Jackson Field Research Station, Jackson, WY; Woodward, D.F., USGS - Jackson Field Research Station, Jackson, WY; Skaar, D., Montana Division of Fish, Wildlife, and Parks, Helena, MT; Brumbaugh, W., USGS - Environmental and Contaminants Research Center, Columbia, MO. The Boulder River and some of its tributaries receive direct effluent from abandoned mine adits and runoff from old tailings piles located in the basin. This assessment identified the biological pathway of metals as measured by concentrations of As, Cd, Cu, Pb, and Zn in biofilm (abiotic and biotic material on rock surfaces), invertebrates, and fish collected from the Boulder River Watershed. Fish tissue samples were also collected for measurements of lipid peroxidation and metallothionein to determine fish health. Increased lipid peroxidation indicates tissue damage, and metallothionein induction has been associated with reduced growth in fish. Fish population estimates were calculated with multiple-pass depletion and mark/recapture methods. The concentrations of metals were greatest in the tributaries where historical mining had occurred and, were often 30x to 100x greater than the concentrations measured in samples collected from reference streams. However, the concentrations of metals in biofilm and invertebrates remained elevated in the mainstem of the river and contained 3x to 10x greater concentrations than reference samples. Whole bodies of fish had elevated concentrations of all metals but, Cd, Cu, and Pb were the metals most concentrated in liver and gill. In fact, the fish gills accumulated similar concentrations of Cd as did the invertebrates. The elevated concentrations of metals were associated with depressed populations and sizes of fish. For example, Cataract Creek had the smallest estimated fish populations, smallest sized fish, and the greatest metal concentrations.

489 Pollution and Predator/Prey Behavior: Links Between the Organism and Both Lower and Higher Levels of Biological Organization. Weis, J.S., Rutgers University, Newark NJ., Smith, G.M. Rutgers University, Newark NJ, Santiago, C. Rutgers University, New Brunswick, NJ. Effects of neurotoxicants on predator/prey behavior can provide clear linkages of effects at different levels of organization from the biochemical to the population and community levels. Many toxicants can affect the nervous system in both developing organisms and adults. These contaminants can alter neurotransmitters,which, in turn can affect behavior. In the case we have studied, mummichogs (*Fundulus heteroclitus*) from a contaminated tidal creek in northern NJ have been previously found to have reduced growth and longevity compared to conspecifics from clean areas. Their activity level is reduced and they are poor at capturing live prey (grass shrimp, *Palaemonetes pugio*) in laboratory tests. They also have reduced levels of serotonin in their brains, which is associated with their lower activity. Fish captured from the field have been found to eat a greater amount of non-nutritious detritus, which may contribute to their reduced growth, condition, and longevity. In lab studies, they are also less able to avoid predation by blue crabs (*Callinectes sapiduss*), which may also contribute to their reduced life span and affect the size-structure of the population. Grass shrimp from this contaminated site, however, do not appear to be impaired in predator avoidance behavior, and are more numerous and generally larger in size than those from the reference site, possibly due to reduced predation by mummichogs.

490 the Cost of Tolerance: a Mechanistic Evaluation Across Levels of Biological Organization. Clements, W.H.*, L.A. Courtney, E.A. Harrahy, N. DuTeau, and P.M. Kiffney. Colorado State University, Fort Collins, CO. We employed molecular genetic, population, and community-level studies to examine the effects of novel stressors (acidification, UVB radiation, and stonefly predation) on organisms collected from a metal-polluted stream. To investigate tolerance of benthic communities to heavy metals, we compared the effects of a mixture of Cd, Cu, and Zn on organisms collected from a metal-polluted stream (the Arkansas River) and an unpolluted reference stream (the Cache la Poudre River). As expected, mayflies (Ephemeroptera) from the Arkansas River were more tolerant of heavy metals than organisms from the Cache la Poudre River. Results of population genetics studies using RAPD-PCR also showed reduced genetic diversity in mayfly populations (*Baetis tricaudatus*) from metal-polluted stations. To examine the cost of tolerance to metals, benthic macroinvertebrate communities collected from metal-polluted and reference streams were exposed to novel biotic (predation) and abiotic (acidification, UVB radiation) stressors. Results indicated that mayfly assemblages from the metal-polluted stream were more sensitive to these novel stressors. Our findings suggest that biotic and abiotic factors interact to structure benthic communities in metal-polluted streams. Ecosystems disturbed by contaminants, particularly those that have been polluted for relatively long periods of time, provide excellent opportunities to study interactions between biotic and abiotic factors.

491 Toxic Contaminants of Human Health Concern in San Francisco Bay Fish: Mercury, PCBs, Organochlorine Pesticides, and Dioxins. Davis, J.A.*, San Francisco Estuary Institute, R. Fairey, C. Roberts, C., Moss Landing Marine Laboratories, G. Ichikawa, California Department of Fish and Game, M. Stoelting and J. Becker, U.C. Santa Cruz, M.X. Petreas, California Department of Toxic Substances Control, and K.T. Taberski, San Francisco Bay Regional Water Quality Control Board. A 1994 study indicated that mercury, PCBs, DDT, dieldrin, chlordanes, and dioxins were of potential health concern for people consuming Bay-caught fish. As a result of this study an interim health advisory was issued for consumption of Bay fish. In 1997 the Regional Monitoring Program for Trace Substances in the San Francisco Estuary began monitoring fish contamination in the Bay. The objectives of this monitoring element are: 1) to produce information needed for updating human health advisories and conducting human health risk assessments and 2) to track trends in contaminant levels in fish over time. Species and tissues analyzed in 1997 included white croaker (muscle and skin), shiner surfperch (whole body), jacksmelt (whole body), striped bass (muscle), leopard shark (muscle), California halibut (muscle), and white sturgeon (muscle). Analytes included mercury, selenium, PCBs (including coplanar PCBs in some samples), organochlorine pesticides, dioxins, and dibenzofurans. A total of 503 fish were collected to yield 71 composite samples and 40 individual fish samples for chemical analysis. Mean wet weight mercury concentrations for each species were: leopard shark (0.905 ppm), striped bass (0.478 ppm), California halibut (0.292 ppm), white sturgeon (0.278 ppm), white croaker (0.213 ppm), shiner surfperch (0.116 ppm), and jacksmelt (0.106 ppm). In general these concentrations were similar to those measured in 1994, but the 1997 striped bass mean was significantly higher than the 1994 mean (0.272 ppm). Data for other analytes are also summarized.

492 Linking Migration Behavior of Hudson River Striped Bass with Congener Specific PCB Patterns. J.T.F. Ashley, J.E. Baker, S.Q. Wales, E. Zlokovitz, and D. Secor. Chesapeake Biological Laboratory, University of Maryland, Solomons, MD. Since 1976, the commercial striped bass fishery has been closed due to PCB concentrations in edible portions exceeding the FDA's advisory level of 2 μg/g-wet weight. Forty fish, collected throughout the Hudson River subestuary, were analyzed for congener specific PCBs. Life-time migration behaviors were determined by otolith analysis. Total PCB (*t*-PCB) concentrations in striped bass were positively correlated with river mile. Moreover, through otolith chemistry, mean salinity encountered during the two most recent growth seasons prior to capture was inversely correlated with total PCB body burden ($r=-0.71$). Striped bass residing in freshwater and oligohaline portions of the estuary had higher *t*-PCB levels. To further substantiate the link between migratory behavior and PCB accumulation, differences and similarities in the congener specific PCB patterns from fish were examined. Those fish residing in the upper estuary near the source had PCB patterns dominated by lighter chlorinated congeners (di-, tri-, and tetrachlorobiphenyls). Conversely, those fish spending the majority of their life in more saline waters of the estuary or those migrating frequently from the upper estuary to the lower, had patterns dominated by more heavily chlorinated congeners. Fish collected from the New York Harbor (NYH) area also had patterns dominated by lighter chlorinated congeners, suggesting that in addition to the upper estuary, NYH may be a source of unweathered PCBs which are available for bioaccumulation.

493 Bio-accumulation and Biomagnification of Persistent Organochlorine Contaminants in the New Zealand Marine Food Web. Day, P.J., Castle P., Victoria University of Wellington, Wellington, NEW ZEALAND; Leathem, S.V. Jones, P.D.* ESR (Institute of Environmental Science and Research), Lower Hutt, NEW ZEALAND. Marine mammals have been shown to accumulate significant concentrations of persistent organic pollutants (POPs) even in relatively pristine environments. To investigate this accumulation in the relatively uncontaminated environment of New Zealand we have analysed blubber samples from New Zealand marine mammals and their food chain as well as coastal sediment samples from the vicinity of the largest adjacent population centre. Marine mammal species analysed include the New Zealand fur seal (*Arctocephalus forsteri*) and Hector's dolphin (*Cephalorhynchus hectori*). A range of biota from different trophic levels were also analysed. Analytes measured were 25 polychlorinated biphenyl (PCB) congeners, including non- and mono-ortho substituted congeners, and a range of organochlorine pesticides. The concentrations of POPs in sediment and non-marine mammal biota samples were considerably (> 2 orders of magnitude) lower than in similar northern hemisphere studies while concentrations in marine mammals were generally within 1 order of magnitude of northern hemisphere concentrations. As a result of these concentrations calculated biomagnification factors (BMFs) suggest greater biomagnification of contaminants in the New Zealand marine environment, particularly in Hector's dolphin, compared to northern hemisphere studies.

494 **Organochlorine Accumulation within the Food Web of Grand Traverse Bay, Lake Michigan: Investigating Current Sources.** Stapleton, H.M., Jeremiason, J.D. , Baker, J.E., Chesapeake Biological Laboratory, University of Maryland, Solomons, MD, Ostrom, P.,Michigan State University, Lansing, MI. The food web of Grand Traverse Bay, Lake Michigan was sampled seasonally from April through September 1997 as part of a study to examine the influence of atmospheric deposition and sediment recycling and resuspension as sources of organochlorine contaminants, and in particular Polychlorinated Biphenyls (PCBs) to its fishery. Samples were composited according to month and size class, and analyzed for total PCB concentrations and organochlorine pesticides. The representative species sampled include: Lake Trout (*Salvelinus namaycush*), Bloater (*Coregonus hoyi*), Alewife (*Alosa psuedoharengus*), deepwater sculpin (*Myoxocephalus thompsoni*), whitefish (*Coregonus clupeaformis*), mysid shrimp (*Mysis relicta*), and amphipods (*Diaporeia hoyi*). The total PCB burdens ranged from a high of 1584 ng/g-wet in Lake Trout to 41 ng/g in deepwater sculpin within the fish sampled. Seasonal variations in total PCB contaminant burdens were only observed in alewife composites and a significant correlation between size class and PCB burdens were observed in deepwater sculpin. Isotopic analyses using $\delta^{15}N$, suggest that deepwater sculpin are at a higher trophic position than alewife and bloater which is not consistent with PCB accumulation trends observed in the other fish sampled. Fingerprint analyses on PCB patterns within the fish sampled suggest that deepwater sculpin may be exposed to PCBs through a different route than are the pelagic species, perhaps indicating sediment sources.

496 **Pesticidal Contamination of Non-fatty Food Commodities in India.** Arora, R.K.*, SKUAST, Udheywalla, Jammu Tawi, India; Jaglan, R.S., Dept. Entomol., CCS HAU, Hisar, India. The pesticidal contamination of environment(soil, water and food commodities) has been maximum in developing countries. This has been due to indiscriminate and careless use of pesticides by illiterate farm workers. So far, only 143 pesticides stand registered under the insecticide act, 1968 on regular basis for their import, manufacture and use in India. Inspite of 20 pesticides banned including DDT and BHC, India tops in percent incidence of contamination of food commodities mainly with DDT and BHC. About 60 percent of the non-fatty food commodities have been found to contain residues of organochlorine pesticides and 20-25 per cent above maximum residue limits and acceptable daily intake fixed under the Prevention of Food and Adulteration Act, 1954 by Ministry of Health, Govt. of India. As a result of high level of contamination of food commodities, dietary intake of DDT and BHC are maximum in indian population. Accumulation of DDT and BHC in human adipose tissues is also common in India. Some kitchen processes have been suggested to bring down residues by 20-100 per cent. Biotechnology based upon microbial and molecular genetic techniques including cell selection, embryo rescue, protoplast fusion and recombinant DNA technology may help in developing resistant varieties to different pests in the years to come to minimise use of pesticides. Presently, to ensure minimum contamination of food commodities and maximum safety to environment, selective and judicious use of pesticides have been suggested as a component of integrated pest management. Besides, regular sampling of food commodities for pesticides residue content and consideration of health costs while deciding pesticide registration and use in India are some pre-eminent strategies outlined.

497 **High Chromium Levels in Biota from Shipyard Creek, South Carolina: A Chromium Hot Spot.** Hyland, J.L.*, NOAA, Charleston, SC; Boothe, P.N., Texas A&M University, College Station, TX. A study of chromium contamination in edible tissues of fish and shellfish was conducted during the summer 1997 in Shipyard Creek, S.C. as part of the Environmental Monitoring and Assessment Program (EMAP) in the Carolinian Province (Cape Henry, VA - St. Lucie Inlet, FL). Shipyard Creek is a small tributary of the lower Cooper River in the vicinity of Charleston Harbor. Chromium in the sediments of this estuary has been found at concentrations up to 21,000 ppm (dry weight), which exceeds the ER-M sediment quality guideline for chromium by a factor of 56. Over 100 samples of spot, croaker, oysters, blue crab, and penaeid shrimp were collected from Shipyard Creek and analyzed for chromium by instrumental neutron activation analysis (INAA). Wet-weight concentrations in these samples ranged from 0.17 - 1.1 $\mu g/g$ for fish, 2.7 - 34.3 $\mu g/g$ for oysters, 0.14 - 24.3 $\mu g/g$ for blue crabs, and 7.5 - 77.8 $\mu g/g$ for shrimp. Chromium concentrations in all four species were significantly higher in Shipyard Creek samples than in samples from other sites. Differences were the strongest for the three shellfish species. Moreover, a large percentage of the Shipyard Creek samples - 95% of the shrimp, 26% of the crabs, and 15% of the oysters - had chromium concentrations that exceeded FDA's human-health "Levels of Concern" (13 $\mu g/g$ for mollusks and 12 $\mu g/g$ for crustaceans). In contrast, none of the samples from sites outside Shipyard Creek contained chromium in excess of these guidelines. These data suggest a potential risk of contaminant exposure to humans from the consumption of seafood from this estuary.

498 **Trophic Transfer of Metals Leached from Pressure-treated Wood.** Weis, P.*, UMDNJ-N.J. Medical School, Newark, NJ; Weis, J.S., Rutgers Univ., Newark, NJ. Metals leached from chromated copper arsenate (CCA)-treated wood, taken up by epibiota and trophically transferred to their motile consumers, were measured in organisms caged with treated and untreated panels which had been submerged in an estuary for up to three months. Epibiota on the treated wood panels had more copper and arsenic than epibiota on untreated wood panels, and amphipods living on the treated panels had more copper. However, grass shrimp (*Palaemonetes pugio*) and two teleost fish, the naked goby (*Gobiosoma bosci*) and mummichog (*Fundulus heteroclitus*), in the cages with treated wood had metal levels comparable to those caged with untreated panels. Thus, trophic transfer was not demonstrated, suggesting that the treated wood was not presenting a hazard at higher trophic levels. There was no evidence of biomagnification among the consumers. In earlier studies of trophic transfer of CCA leachates, two gastropod species had adverse effects from feeding on epibiota, but two fish species did not. Fish may have more efficient mechanisms for regulating metal levels in their tissues. (Supported by NOAA Office of Sea Grant)

499 **The Influence of Animal Size and Exposure History on the Subcellular Partitioning of Cd and Zn in the Bivalve *Potamocorbula Amurensis* and the Implications for Metal Trophic Transfer.** Wallace, W.G.*, Brown, C.L. and Luoma, S.N., United States Geological Survey, Menlo Park, CA 94025. Recent studies demonstrate that metal trophic transfer may be predicted based on subcellular compartmentalization within prey and that it is the metal associated with the cytosol and intracellular organelles which is the most bioavailable to predators. Consequently, factors influencing metal partitioning may affect metal trophic transfer. The

goal of the present study is to investigate how animal size and exposure history influence subcellular partitioning of Cd and Zn in *P. amurensis*. A strong size dependence in subcellular metal partitioning appears to result from size dependent metal detoxification. Small clams had 10% Cd bound to metallothionein (MT) proteins and 2% Zn stored in metal-rich granules (MRG). Large clams had 35% Cd associated with MT and 16% Zn sequestered via MRG. Because of differences in bioavailability to predators of this detoxified metal (Cd-MT, Zn-MRG), it is estimated that when feeding on small vs. large clams Cd bioavailability would increase from 56% to 64% and that of Zn would decrease from 80% to 70%. Additionally, exposure history was also found to have an important role in controlling these processes. Clams from an historically contaminated site in San Francisco Bay had 18% more Cd associated with the biologically available fraction than clams from a relatively clean site. There was a similar relationship for Zn, though the difference was greater (26%). These site specific differences in biologically available metal, in conjunction with differences in total metal body burdens translate into a 3.0x greater Cd exposure and a 2.0x greater Zn exposure for predators ingesting clams from the contaminated versus the clean site.

500 ***In situ* and Laboratory Experiments to Determine the Importance of Food as a Cadmium Source to the Aquatic Insect *Chaoborus*.** Munger, C.*, Hare, L., and Tessier, A., INRS-Eau, Université du Québec, Sainte-Foy, Québec, Canada. Metals can be accumulated by aquatic animals from the surrounding water or from the food that they eat. The relative contribution of water and food as uptake routes is poorly known, yet such information is important if we are to measure metal movements in food webs and to construct reliable models of metal accumulation in animals. In the laboratory, we showed that food (zooplankton) was the major Cd source for larvae of the aquatic insect *Chaoborus*. We tested our finding in the field. *Chaoborus* from an uncontaminated lake were transferred to water-permeable mesocosms in a Cd-contaminated lake where the predator was given access to various quantities of natural Cd-rich food (zooplankton) collected from the surrounding water. Both larval mass and larval Cd content increased with increasing quantities of Cd-rich food offered, supporting our hypothesis that food is the insect' major Cd source in nature. Having demonstrated the importance of Cd taken up in food, we measured the influence of ingestion rate and prey type on Cd accumulation by the predator. Larvae from an uncontaminated lake were offered various quantities of two types of prey collected from a Cd-contaminated lake. Our results suggest that Cd accumulation by the predator was dependent on its ingestion rate but independent of the type of prey it consumed. We suggest that differences in prey abundances among lakes could explain in part the reported lake to lake variation in predator Cd concentrations.

501 **Environmental Transport and Fate of PCBs in Lotic Ecosystems.** W.A. Robison*, US FWS, Cookeville, TN, W.J. Birge and D.J. Price. University of Kentucky, Lexington. KY. Transport and distribution of PCBs were studied in three Kentucky stream systems ranging in size from second to fifth order. Point source releases of PCBs in each stream system resulted in state agencies issuing fish consumption advisories. Water, sediment, floodplain soil and fish-tissues were analyzed for PCBs. Longear sunfish, green sunfish, stonerollers and banded sculpins were among the species analyzed and represented different trophic levels/feeding habits. In green sunfish, relatively rapid disappearance of PCB residues was noted, and may have better indicated current PCB bioavailability in each system. In one stream system, PCBs were detected in water, sediment and floodplain soil sixty-five miles from the known source. Mean concentrations of Aroclor 1248 in sediment decreased from 280 ppm near the source to 0.09-0.12 ppm about 100 mi. downstream. Although contaminated groundwater may have contributed substantial stream PCB loading near sources, sequential resuspension of PCB-contaminated sediment was considered the main long-range transport mechanism. Transport reduction factors were greatest for stream sediment. While flood events may have distributed PCBs over 20,000 acres of floodplain soil, possibly affecting background concentrations in these areas, the greatest concentrations of PCBs were present in close proximity to the industrial PCB source. About 50 percent of fish collected from the Mud River contained residues greater than the FDA action level (2.0 ppm) in edible portions. Fish collected within 4 mi. of the source contained PCBs ranging from 9.4 to 33.3 mg/kg. Estimated PCB bioconcentration factors (BCFs) ranged up to 300,000 based on water and fish-tissue concentrations. BCFs decreased with downstream distance from the source, possibly indicating reduced PCB bioavailability. Relationships between various tissue concentrations were also examined. These results indicated the potential for persistence, continued sublethal effects, and ecological risk of PCBs in lotic systems. Consideration has also been given to potential innovative remediation or restoration techniques.

502 **Pre- and Post- Remediation Concentrations of Chlorinated Hydrocarbons in Spotted Seatrout and Blue Catfish of the Calcasieu River Estuary, Louisiana.** Tourtellotte, G.H.*, QST Environmental, Tampa, Fl; Marsh, W.T., QST Environmental, Gainesville, Fl; Wood, M. S., PPG Industries, Lake Charles, La. PPG Industries, Inc. is conducting a quarterly biomonitoring program, started in 1989, to determine concentrations of the chlorinated hydrocarbons, hexachlorobenzene (HCB) and hexachlorobutadiene (HCBD), in estuarine biota near its Lake Charles, Louisiana facility. The overall goal of the study is to determine if mitigative measures in the form of industrial process changes, the construction of a bypass canal, and the isolation of an old discharge canal result in declines of the tissue burdens of HCB and HCBD in estuarine biota important as recreational and commercial fisheries. Two fish species, spotted seatrout (*Cynoscion nebulosus*) and blue catfish (*Ictalurus furcatus*), utilize the study area seasonally in response to the seasonally variable salinity structure of the estuary. Spotted seatrout occur primarily during seasons of high salinity (summer and fall), and blue catfish occur primarily during seasons of low salinity (winter and spring). Five years of pre-remediation monitoring and four years of post-remediation monitoring have shown that remediation programs instituted by PPG Industries have significantly reduced the concentrations of HCB and HCBD in the edible tissues of these two fish. Mean concentrations of HCB in spotted seatrout have been reduced from 39 ppb to 17 ppb. Mean HCBD concentrations in spotted seatrout have been reduced from 213 ppb to 114 ppb. Blue catfish mean HCB concentrations have been reduced from 106 ppb to 5 ppb, and blue catfish mean HCBD concentrations have been reduced from 1,087 ppb to 55 ppb.

503 **Response of Hudson River Estuary-New York Harbor Sediments and Striped Bass to Multiple Sources of PCBs.** Farley, K.J.*, Thomann, R.V., Manhattan College, Riverdale NY. PCB concentrations in sediments and striped bass in the Hudson River Estuary-New York Harbor have remained elevated throughout the 1990's largely due to continuing sources of PCBs from the General Electric plant site on the upper Hudson River, wastewater treatment plants and combined sewer overflows from the New York metropolitan area, and atmospheric deposition. The objective of our study was to determine the overall transport, fate, and bioaccumulation of PCBs in the system and to evaluate the relative impacts of various sources on PCB contamination levels. For this purpose, we constructed a fully time-variable, congener-specific model for the Hudson River Estuary-New York Harbor ecosystem and used the model to analyze PCB water, sediment, and fish congener data from EPA Region II, NOAA, and GE. From analysis of field data and modeling results, we have found that PCB loads from the GE plant site (which have varied from 0.5 to 15 kg/day) are largely carried downstream toward New York Harbor during Spring high flow events. During the remainder of the year, however, PCBs are largely lost from the river by volatilization. On an annual basis, transported loads from the upper Hudson have been found to be comparable to the 0.4 kg/day of PCBs from sources in the New York metropolitan area. Based on comparisons of calculated and observed congener distributions, PCB losses by biodegradation, dechlorination, or fish metabolism are not significant in the estuary. Variations in striped bass migration patterns, however, play an important role in determining PCB bioaccumulation.

504 Establishing a Consistent Quantitation Basis for Long-term Modeling of PCBs in the Hudson River. Butcher, J.B., Tetra Tech, Research Triangle Park, NC; Garvey, E.A.*, TAMS Consultants, Bloomfield, NJ. PCB contamination of water, sediment, and biota in the Hudson River has been studied since the early 1970's. A Reassessment Remedial Investigation/Feasibility Study is currently underway, including both new data and a retrospective analysis of historic data. With 25 years of monitoring, the Hudson River offers a unique opportunity for calibration and testing of long-term models of PCB transport and bioaccumulation in a flowing river. Fitting such a model, however, requires the integration of PCB data analyzed under a wide variety of protocols. Recent data have primarily been analyzed for individual congeners using capillary column gas chromotagraphy. Historical data for water, sediment, macroinvertebrates, and fish, however, are primarily by packed column methods analyzed against Aroclor standards and reported either as Aroclor equivalents or total PCBs. Further, quantitation methods for historical data varied from Webb and McCall-type partitioning of the entire chromatogram to a variety of schemes based on calibration against a limited number of packed-column peaks. Most of the original packed-column chromatograms are lost. The objective of this study was to examine the historical quantitation methods for Hudson River media and evaluate what would have been reported relative to a congener-based analysis. This yields a consistent common-denominator interpretation of historical and modern PCB analyses suitable for fitting long-term models. Our results indicate that the historic methods are not equivalent to the total sum of PCB congeners. However, after introducing appropriate corrections, the historical packed-column results can be converted to a good approximation of the sum of trichloro and higher-chlorinated biphenyls, yielding a consistent quantitation basis. We also conclude that the historic data are largely uninformative as to mono/dichlorobiphenyl concentrations.

505 Development of a Probabilistic Depth of Scour Model For PCB-Contaminated Cohesive Sediments In The Hudson River. Dilks, D. W.*; Helfand, J. S.; Verhoff, S. W.; Raghunathan, R. K., Limno-Tech, Inc., Ann Arbor, MI. The objective of this investigation is the development of a probabilistic model capable of predicting the depth of scour of cohesive sediments in Thompson Island Pool of the Hudson River, caused by flood events. Activities consisted of application of a 2-dimensional hydrodynamic model predicting bottom shear stresses, and development of the probabilistic submodel that predicts the scour depth based upon applied shear stress. The hydrodynamic model RMA-2V was applied using available stream channel and floodplain morphometry data, and calibrated to historical river stage and velocity data. Previous studies provided the data necessary for the creation and implementation of the probabilistic submodel; these included 1) A deterministic formulation to predict cohesive sediment erosion as a function of shear stress, 2) Field data characterizing sediment properties and PCB contamination in the study area. 3) Laboratory and field studies of the erosion properties of upper Hudson River cohesive sediments. The erosion equation was linearized to predict erosion from a dimensionless shear stress parameter and a lumped parameter representing site-specific sediment properties. A regression was performed using the linearized model relating observed erosion to shear stress. The regression statistics were then used to define the uncertainty in predicted scour and regression coefficients. By combining PCB depth profile data with the depth of scour predictions, PCB release from cohesive sediments in response future to flood events was estimated. Model results indicate that the depth layers of highest PCB contamination will not be exposed for the 100 year flood condition. The model improves upon previous formulations in that model uncertainty is defined from actual data taken at the study location.

506 Evidence for PCB Loss from the Sediments of the Upper Hudson River. Garvey, E. A.*, Hunt, C., TAMS Consultants, Inc., Bloomfield, NJ; Butcher, J. B., Tetra Tech, Research Triangle Park, NC. Hudson River PCB contamination dates back to the early 1950s with the onset of PCB-contaminated discharges from two General Electric facilities located near Ft. Edward, NY. As part of the ongoing USEPA reassessment of PCB contamination in the Hudson, current estimates of sediment PCB inventories were obtained in 1994 from 170 sediment cores, principally collected from PCB *hot spots*. Previously, between 1976 and 1984, two major surveys of sediment PCB contamination were conducted by the NYS Department of Environmental Conservation (NYSDEC). A comparison of the current and historical sediment PCB inventories was made to integrate the effects of PCB release from the sediments to the water column, continued PCB release from the General Electric facilities, PCB dechlorination, and sediment deposition and transport during the intervening period. Additionally, it was unclear whether the earlier, more extensive surveys would still be useful in estimating current PCB inventories in areas not surveyed in 1994. After converting the historical PCB data to a basis consistent with the current analytical scheme, the estimates of sediment PCB inventories from the current and historical surveys were compared. Results showed a marked decline in sediment PCB inventories in the historically most contaminated locations, declines which were well beyond that which could be explained by dechlorination processes alone. When the inventories were examined on a molar basis, the evidence strongly suggested PCB loss from the sediment to the water column as the principal means for the decline. This conclusion was based on the fact that dechlorination processes preserve PCB molecules while causing PCB mass loss and that the sediment PCB molar inventories had declined an average of 30 percent.

507 Reductive Dechlorination of PCBs in Lake Hartwell, SC, USA. Pakdeesusuk, U.*; Lee, C. M.; Coates, J. T.; Woolfolk, C. T.; Elzerman, A. W.; Freedman, D. L., Environmental Engineering and Science, Clemson University, Clemson, SC, 29634-0919. Evidence is discussed for microbially mediated reductive dechlorination of polychlorinated biphenyls (PCBs) in the sediments of Lake Hartwell, a US Army Corps of Engineers flood control reservoir located near the point where the Tugaloo and Seneca rivers form the Savannah River. PCB-containing effluents were discharged by a capacitor manufacturer from 1958 to 1977 to a tributary of Twelve Mile Creek which drains into Lake Hartwell. Congener-specific analysis of sediment cores collected in 1987, 1994, and 1998 indicates the accumulation of lower chlorinated congeners and terminal dechlorination products with depth in Lake Hartwell near the entrance of Twelve Mile Creek. Analysis of the cores from sampling locations further downstream provide mixed results indicating that physiochemical weathering processes may be favored over biochemical weathering. Decreases of non-ortho chlorine atoms with depth in the Twelve Mile Creek area of the lake correspond to sections of relatively high concentrations of PCBs in the cores. Increasing concentrations with depth of the dichlorinated PCBs in the 2,2'-/2,6- substitution group and the trichlorinated PCBs in the 2,2',6- substitution group are likely indications of reductive dechlorination in the Twelve Mile Creek area of Lake Hartwell. Although, overall concentrations of PCBs are shown to decline with time, surfacial sediment concentrations of PCBs show upstream sediments continue to contribute PCB contamination to downstream sediments.

508 Enantioselective Degradation of Polychlorinated Biphenyl Atropisomers. Wong, C.S.* and Garrison, A.W., U.S. Environmental Protection Agency, National Exposure Research Laboratory, Ecosystems Research Division, Athens GA 30605-2700. Polychlorinated biphenyls (PCBs) are ubiquitous contaminants that can be biotransformed in some sediments and biota (e.g., via reductive dechlorination). To this date, little research has been done to determine the occurrence, distribution, and fate of the 19 chiral PCB atropisomers (rotationally stable axial enantiomers). The two enantiomers of each atropisomer are expected to have different biological and toxicological properties, and differences in enantiomeric ratios (ERs) of PCB atropisomers may serve as tracers of biotransformation activity in the environment. The enantioselectivity of PCB degradation was investigated to evaluate the applicability of PCB ERs in determining biotransformation. Laboratory studies to determine the enantioselectivity of PCB atropisomer degradation via microbial reductive dechlorination and enzymatic phytodegradation were investigated, and field measurements of atropisomer ERs for a suite of highly contaminated sediments and biotic tissues (e.g., fish, birds, bivalves) were obtained. Preliminary analysis suggests that biota contain non-racemic ERs for several atropisomers, indicative of an enantioselective biological process taking place either *in vivo* or in the organism's environment followed by subsequent bioaccumulation.

509 Effects of the 1997 Red River Flood on Contaminant Transport and Accumulation in the Aquatic Food Chain of Lake Winnipeg. Stewart, A.R.*, Stern, G.A., Lockhart, W.L., Billeck, B.N., Grift, N.P., and Stainton, M.P., Freshwater Institute, Winnipeg, Manitoba. In 1997, the Red River valley experienced the worst flood in over 100 years. Large areas of agricultural and rural and urban land along the Red River extending from Grand Forks, North Dakota to Winnipeg were completely covered in water. Numerous sources of contamination from the flooded areas were identified including agricultural chemicals (insecticides and herbicides), household and commercial hazardous wastes, home fuel-oil heating tanks, preserved wood structures (pentachlorophenol), and hazardous waste disposal sites (toxaphene). A study sponsored by the International Joint Commission (IJC) is being conducted to evaluate some of the environmental impacts of the flood, particularly the transport, fate and bioaccumulation of persistent contaminants (organochlorine pesticides, herbicides, hydrocarbons and metals). Contaminants were measured in water and suspended sediment sampled during the flood in the Red River at points south of Winnipeg (upstream) and at Selkirk (downstream) near Winnipeg and on the major tributary west of Winnipeg. This sampling design allows calculation of the total flux of materials to Lake Winnipeg and also allows isolation of the contributions from the city and from higher in the watershed. Many persistent contaminants are associated with carbon and preliminary results for suspended carbon (3,228,274 kg/day, May 1, 1997) suggest a substantial daily loading of contaminants into the south basin of Lake Winnipeg. In the spring of 1998 samples of water, surficial sediments, benthic invertebrates, and fish were collected from 33 sites in the south basin of Lake Winnipeg and analyzed for contaminants. Relationships between contaminant persistence and bioaccumulation in the Lake Winnipeg food chain will be discussed in terms of the IJC objective to identify environmentally hazardous or sensitive features of the Red River basin to future flooding.

510 Organochlorine Contamination on National Wildlife Refuges in the Lower Mississippi River Ecosystem. Hofelt, C.S.*, Shea, D., N.C. State University, Raleigh, NC; Kelly, J.R., US EPA, Duluth, MN. As part of an ecosystem-level contaminants study for the U.S. Fish and Wildlife Service, we analyzed sediment, fish, and SPMDs for organochlorine pesticides (OCPs) at 26 National Wildlife Refuges (NWRs) in the lower Mississippi River ecosystem. We report on the spatial distribution of OCPs, how concentrations have changed over the last 20 years, and the implications for human and ecological health. We found that OCP concentrations in sediment were uniformly low throughout the region. For example, dry weight concentrations of total DDTs generally ranged from <0.1 to 10 ng/g, though concentrations approached 100 ng/g at Panther Swamp, Tensas River, and Yazoo NWRs. Concentrations of OCPs were much higher in fish and SPMDs, with total DDTs ranging from 30 to 6,000 ng/g in fish and 20 to 2,000 ng/SPMD. The highest concentrations in SPMDs and fish generally followed the pattern in sediments, however some sites were very high in fish but low in sediment and SPMDs. Areas with the highest concentrations tended to have the highest historical use of OCPs and the longest water residence times. The concentrations we found in sediment and fish are generally more than an order of magnitude lower than those reported just 10 years ago. Mean concentrations of OCPs in water were estimated from the SPMD residues and published SPMD sampling rates. These estimates approached 10 ng/L for total DDTs in some samples, which is much higher than we expected or has been reported previously. A simple comparison of our data to established sediment, water, and fish residue guidelines indicates that sediment generally meets most guidelines, but water and fish approach or exceed guidelines at several sites.

511 Lead Shot and the Environment: Going Beyond Wetlands. Thomas, V.G., Dept. of Zoology, University of Guelph, Guelph, Ontario. Lead in its various forms has long been recognized as toxic to human and wild life. A general lead toxic syndrome exists, whose signs vary little, especially among vertebrate species. The toxicity posed by spent lead shot is just a subset of this general syndrome. Effective substitutes for lead shot, derived from either iron, tin, bismuth, or tungsten, have been developed and are marketed in a small number of countries. However,only a mall proportion of those nations which report lead shot toxicosis in their wild life have either considered or begun remediation involving phase-out of lead shot use. Lead shot toxicosis is associated mainly with waterfowl and their predators, which explains why most nations have regulated lead shot use only for wetland hunting. Many tons of lead shot are discharged by hunters over upland sites each year. Sporting clay target shooters deposit an even greater tonnage over both upland and wetland sites each year. Those shot also cause lead toxicosis of wild life and long-term contamination of soil, surface water, and ground water. The regulation of the use of lead shot should extend to all forms of shooting activities as it has in Denmark and The Netherlands. The ranges of migratory birds often covers several nations where lead toxicosis is prevalent, as in Western Europe-Scandinavia, and North-Central America. Requiring the use of non-toxic shot in only one, or a minority, of jurisdictions is not effective remediation, especially if the wintering grounds which harbour large concentrations of birds continue to receive spent lead shot. Successful remediation of lead toxicosis requires securing the entire flyway. Secondary lead poisoning of human beings , especially native people who consume large amounts of wild waterfowl, is a significant toxic risk, and constitutes yet another reason to require the use of lead-free shot for hunting.

512 Sixteen Years of Lead Poisoning in Eagles, 1980-1995: an Epizootiologic View. Kramer, J.L. and P.T. Redig*. The Raptor Center, St. Paul, Minnesota. A 16y (1980-1995) retrospective study was conducted to assess differences in the prevalence of lead poisoning (Pbtx) in Bald (Haliaeetus leucocephalus') and Golden (Aquila chrysaetos') Eagles admitted to The Raptor Center at the University of Minnesota. These years encompass the period before and after federal legislation was enacted restricting the use of lead shot for hunting waterfowl on federal lands (1991). Additional data from 1996 through 1998 has been added to the database without fundamentally changing the conclusions. Of 654 eagle admissions reviewed, all of whom were analyzed for presence of lead residues at the time of admission, 138 eagles exhibited elevated lead residues and were further evaluated for the following: recovery location (generally the upper Midwest), blood lead concentration, month of admission, radiographic evidence of lead in the ventriculus and primary cause of admission. The prevalence of Pbtx in eagles did not change after 1991, but mean blood lead concentrations of lead in the same population decreased. These finding call into question current theories regarding the sources of lead for eagles (i.e. waterfowl) and and suggest alternative sources. There was a coincidence of the greatest rate of admissions of eagles with lead poisoning and the timing of deer seasons and migration from northern reaches of their range into the area. Lead poisoning is a continuing problem both regionally and internationally, and man variable related to this toxicity have yet to be conclusively defined.

513 Lead Exposure in Bald Eagles and Golden Eagles from the Canadian Prairie Provinces. Wayland, M.*, Environment Canada, Saskatoon, SK, Canada; Bollinger, T., Canadian Co-operative Wildlife Health Centre, Saskatoon, SK, Canada; Scheuhammer, A., National Wildlife Research Centre, Hull, PQ, Canada. Lead poisoning has been recorded in raptors in North America, Europe and Asia. It has been documented most thoroughly for bald eagles (*Haliaeetus leucocephalus*) in the USA and has occurred in golden eagles (*Aquila chrysaetos*) in the USA and Europe. The major source of lead is believed to be lead shot and lead bullet fragments in tissues of game animals. While there is a strong association between ingestion of lead ammunition from various types of prey and lead poisoning in raptors, the importance of lead shot in waterfowl is less certain. It is important to distinguish between waterfowl and other prey as the main sources of lead exposure in raptors because, in several countries, lead shot has been or will soon be regulated for waterfowl hunting but not for other hunting. In this study, we examined lead poisoning and high lead exposure in bald eagles and golden eagles from the Canadian prairie provinces. Approximately 130 dead eagles were autopsied and their liver and/or kidney lead levels were determined. The prevalence of lead poisoning was approximately 12% in both species. Elevated exposure (including poisoning) occurred in 23% of golden eagles and 16% of bald eagles. Lead-exposed bald eagles were found more frequently in areas of high waterfowl hunting than in areas where waterfowl hunting was low. In contrast, lead-exposed golden eagles were found more frequently in areas with low waterfowl hunting. Lead shot in crippled or sick waterfowl may be a major source of lead in bald eagles. Other sources of lead more likely explain high lead exposure in golden eagles.

514 Environmental Behavior and Management of Lead at Shooting Ranges. Byrne, R. L., Wildlife Management Institute, Washington, DC. This paper addresses the environmental mobility of lead in surface water, ground water, sediment and soil, the factors controlling that mobility, and methods of managing lead mobility at shooting ranges. The mobility of lead derived from bullets and shot is controlled by a number of geochemical processes, chief among which are oxidation/reduction, precipitation/dissolution, adsorption/desorption and complexation/chelation. The interaction of these processes with physical processes such as erosion is also important. Site-specific conditions determine which processes and interactions are most important at any particular range. The primary chemical factors influencing lead mobility at shooting ranges are pH, precipitating agents (e.g., phosphates, sulfates, sulfides, carbonates), and sorbents (e.g., clays, organic carbon, iron and manganese oxides and hydroxides). Major influences on lead mobility are exerted by both extrinsic factors such as rainfall quality and quantity, and intrinsic factors such as soil pH or vegetative cover on the range. Natural conditions on some ranges keep lead mobility at a minimum. If lead mobility is a potential concern, intrinsic factors can be managed to accommodate extrinsic factors in an environmental stewardship program to control lead mobility. The most viable options to control lead mobility at shooting ranges include lead recovery/recycling, control of runoff, control of pH with additions of agricultural lime, precipitation via additions of agricultural lime or phosphate, and sorption to clays. Optimum control may require a combination of options selected and applied in response to site-specific geochemical and physical conditions.

515 Use of lead shot for harvesting game animals: Continued deposition of lead into the environment and implications for human consumers of harvested game. Money, S.L.*; Braune, B.M.; Fontaine, A. National Wildlife Research Centre, Hull, Quebec, Canada. Environmental deposition of lead in the form of lead shot used to harvest game in Canada is estimated to be at least 2000 tonnes annually, equaling the estimated releases of lead from industrial sources. In 1997, Environment Canada amended the Migratory Birds Regulations prohibiting the use of lead shot for hunting most migratory game birds nationwide. With the full implementation of this ban in 1999, and continued industrial declines in emissions, upland game hunting and target shooting, which will continue to use lead shot, will represent an increasing proportion of the total amount of lead discharged into the Canadian environment. Fragments of lead shot found in edible tissues of harvested game pose a direct risk of lead exposure to consumers of these tissues, including humans. Although the overall incidence of lead shot retention in humans may be low, there are subpopulations that traditionally harvest and consume above-average quantities of wild game, an activity that may increase their risk of lead exposure.

516 Lead Levels in Mourning Doves from South Carolina: Potential Hazards to Doves and Humans. Burger, J., Rutgers University, Piscataway, NJ; Kennamer, R.A., Savannah River Ecology Laboratory, Aiken, SC; Brisbin, I.L., Jr.*, Savannah River Ecology Laboratory, Aiken, SC; and Gochfeld, M., Rutgers University, Piscataway, NJ. Levels of lead and other environmental contaminants were determined in muscle samples from mourning doves (*Zenaida macroura*) foraging on a contaminated Superfund waste site at the 780-km^2 Department of Energy's Savannah River Site (SRS) near Aiken, South Carolina. These contamination levels were compared to those of doves collected during fall hunts on public dove fields located on lands surrounding the SRS. Lead levels were the highest in muscles of birds collected off of the SRS in fields with prior histories of dove hunting. Levels of radionuclides and all metals including lead were generally within the lower range of those reported in the literature for all birds, and were unlikely to pose health risks to the birds themselves. Lead levels in doves from the public hunting fields were well below reference levels for adult human intake and only would have been a problem if a growing child ate 120 g or more of dove meat every day of the year. It is perhaps ironic that doves from a contaminated waste site of the SRS were less likely to pose a human health risk from lead if eaten, than those from offsite public hunting areas where exposure to spent lead shot apparently has increased lead contamination levels in these birds.

517 Human Health Risk Associated with Environmental Lead Exposure. Patrick Levallois. While lead has been in use since ancient times, its toxic effects are still under study. Effects of high exposure such as encephalopathy, anemia and chronic nepropathy are well known. But knowledge on the effects of chronic low dose exposure has progressed only in the recent years. Cohort studies on low exposure of children demonstrated that blood lead levels in the range of 0.48 - 0.72 μmol/L (10-15 μg/dL) in infants and young children are associated with neurobehavioral and cognitive impairment. No LOAEL has been clearly defined for such an effect and its reversibility is uncertain. Other subtle effects of low dose exposure in adults are still under study : elevation of blood pressure, impairment of renal function and fertility. Acceptable concentrations of lead in blood samples will also be discussed.

518 Ingested and Retained Lead Shot in the Gastrointestinal Tracts of Humans. Reddy, E.R. Memorial University of Newfoundland. Seventy patients with retained lead shot in their gastrointestinal tracts from eating wild game, discovered during routine radiographic studies, were followed for periods ranging from two months to thirteen years. The largest numbers of lead shot accumulated in the appendices. The number varies from 1 to over 200. Twenty patients who had barium studies showed no obstruction of the bowel or any evidence of luminal obstruction of their appendices. Eight patients who had appendectomies showed no evidence of appendicitis on tissue examination. None of the other patients developed any clinical evidence of appendicitis or lead poisoning. It is concluded that no casual relationship exists between ingestion and retention of lead shot, their number, duration of their presence in the gastrointestinal tracts of the humans and appendicitis.

519 The Use of Lead Shotshell for Subsistence Harvesting by First Nation Cree: Human Health Concerns. Tsuji, L.J.S.*, York University, Toronto, Canada; Nieboer, E., McMaster University, Hamilton, Canada; Karagatzides, J., Trent University, Peterborough, Canada; Katapatuk, B., Weeneebayko General Hospital, Moose Factory, Canada. Lead exposure for First Nation Cree of the western James Bay area through ingestion of wild game harvested with lead shot is of concern because we have shown that approximately 15% of randomly selected radiographic charts examined at the regional hospital had evidence of pellets lodged in the digestive system, intraluminally and/or in the appendix. Further, we have found that 9% (33/731) of edible skeletal tissue samples, obtained through the harvesting of wild game with lead shot in the region, had lead levels greater than the Health Canada guideline of 0.5 mg/g ww. Elevated lead levels in these tissues were shown through radiography and subsequent atomic absorption spectrometry (AAS) to be the result of lead pellets/fragments being embedded in the tissues. We will also present evidence illustrating elevated dentine (tooth) lead levels in both adults and children of the region. Adult teeth were collected during the period 1989 to

1995 from patients who needed their tooth extracted for carious or periodontal reasons. One hundred and thirty two teeth were collected from 89 individuals, 54 males and 35 females. Root dentine samples were analyzed by flameless AAS. Dentine lead data were not significantly different for the sexes. Significant differences between several age categories were found and dentine lead levels increased significantly with age. The observed lead levels were found to be comparable in magnitude to those reported in other studies, even though our samples were from individuals living in a remote region. Similarly, elevated lead levels in dentine chips were found for naturally exfoliated primary teeth (15 of 61 teeth had lead levels >10 mg/g dw). When these data are considered as a whole, it appears that lead shot is a major source of lead exposure in subsistence harvesting Cree of the western James Bay region.

520 Lead Poisoning among Inuit Children: Identification of Sources of Exposure. Lévesque, B., Centre de santé publique de Quebec, Quebec, Canada; Dewailly, E., Centre de santé publique de Quebec, Quebec, Canada; Dumas, P., Centre de toxicologie du Quebec, Quebec, Canada; Rhainds, M., Centre de santé publique de Quebec, Quebec, Canada. Since the beginning of the 1980s, lead contamination of the environment has progressively become a major public health preoccupation, mainly as regards to neurobehavioral effects on fetuses and young children. Based on epidemiological evidence, the US Centers for Disease Control (CDC) revised its sanitary intervention threshold to I0μg/dl in 1991. In the province of Quebec (Canada), the results of cord blood studies on lead carried out between 1993 and 1995 showed that 7.6% (n=238) of Inuit newborns from Nunavik had blood lead levels of 10 μg/dI and more, compared to 0.2% (n=955) of babies from the Southern part of Quebec. By reviewing the sources of exposure of Inuit children, it was concluded that nutrition is the most probable source of exposure, especially through two types of food, waterfowl most probably contaminated by lead shots, and caribou contaminated by its lichen-based diet. The isotopic ratio techniques may help to determine distinct sources of lead exposure. We characterized the stable lead isotopes in the blood samples collected from 29 newborns with lead levels equal to or greater than 10 μg/dI. Each of these children was paired with a control from the same community and 3 other controls previously sampled from a study carried in the South of Quebec. For each blood sample three isotopic ratios (206/204, 206/207, 206/208), are calculated. Means and variances of the three groups will be compared. Results and the discussion of these results will be presented.

521 Use of Multi-Media Models for Defining Criteria and Framework for Evaluating Persistence and Long-Range Transport of Chemicals. C.E. Cowan*, The Procter & Gamble Company, Cincinnati, OH, USA; T. F. Parkerton, Exxon Biomedical Sciences Inc., East Millstone, NJ, USA; D. van de Meent, RIVM, Bilthoven, The Netherlands. There are several international and national initiatives in progress to identify persistent organic pollutants (POPs) and in activities to prioritize chemicals for assessment and potential management action. Two of the criteria that are often used to identify these substances are persistence and the ability to undergo long-range transport to remote regions beyond those where the chemical is manufactured and/or used. However, exactly what physical/chemical characteristics of the chemical and associated criteria to use to determine if a chemical is persistent or has the potential for long-range transport is still very much in debate. It is also recognized that these physical/chemical characteristics may vary with the climatic and environmental conditions and that they do not act independently of each other nor of the release pattern (e.g., geographic distribution and environmental media distribution). Therefore, various multi-media models have been used in determining which set of physical/chemical characteristics and associated criteria can be used in the initial screening of chemicals for the potential for persistence and long-range transport. Multi-media models can also be used to look at the linkage between the distribution of releases and the concentrations of the chemicals in remote regions. Thus, the recommended framework for evaluating the potential for a chemical to be persistent and undergo long-range transport includes screening using physical/chemical properties and criteria values as well as use of multi-media models in a more detailed analysis of the potential for transport and estimation of environmental concentrations in remote regions. In this presentation, the final framework, which was the product of a SETAC workshop that brought together an international team of experts to address this problem. will be presented with emphasis on how multi-media models were used and can be used to assess these chemicals.

522 TRIM: A Multimedia, Multipathway Framework for Assessing Human and Ecological Exposure and Risk. Vasu, A.B.*, U.S. EPA, RTP, NC; Hetes, R.G., U.S. EPA, RTP, NC; Palma, T., U.S. EPA, RTP, NC; McKone, T.E., Lawrence Berkeley National Laboratory, Berkeley, CA. In order to meet the requirements of the Clean Air Act of 1990 and to respond to new risk assessment guidance, the EPA must develop tools for evaluating the transport, transformation, and fate of pollutants in multiple media and at multiple spatial and temporal scales. These tools must also address both human and ecological exposure and risk and provide for sensitivity and uncertainty analyses. To meet these needs, the Office of Air Quality Planning and Standards of the U.S. EPA is developing the Total Risk Integrated Methodology (TRIM), a multimedia, multipathway, time-series simulation modeling system for the assessment of human and ecological risks resulting from hazardous and criteria air pollutants. This modeling system is being designed to be scientifically defensible, flexible, and user friendly. TRIM includes a combined set of features that have not been available in a single modeling system. These features include: a modular design which allows flexibility in the construction of multimedia fate models; the capability to vary the spatial and temporal scale for each pollutant and assessment scenario; optimized numerical solution methods; and, methods for explicitly treating uncertainty and variability. The fate and transport model in TRIM uses a dynamic mass-conserving approach to estimate pollutant concentrations received by selected receptors. Of the six modules of TRIM, two modules, the environmental fate and transport module and the exposure-event module, are fully under development. Test cases demonstrating unique features of these modules will be presented.

523 Uncertainty Analysis of Human Toxicity Indicators for LCA. Hertwich, E.G.*, McKone, T.E., Pease, W.S., University of California, Berkeley, CA and Environmental Defense Fund, Oakland, CA. The human toxicity potential (HTP) is used to compare the potential human health impacts of substances listed in life cycle inventories and toxics release inventories. It is the most sophisticated toxicity indicator available today because it includes potential exposure in addition to toxic potency. This presentation focuses on the innovative part of HTP, the use of CalTOX, a multimedia, multiple-pathway exposure model, to calculate potential exposure. We systematically analyze the different types of uncertainty connected to the use of this exposure model. For parameter uncertainty and variability, we use Monte Carlo analysis to construct probability density functions for selected chemicals. We compare this uncertainty to the distribution of exposure values for all chemicals. To evaluate decision rule uncertainty, we identify a number of indicator design questions that could be resolved in various ways and discuss the importance of these design decisions or conduct a sensitivity analysis to evaluate their importance. We identify a number of contributions to model uncertainty and illustrate its impact using sensitivity analysis. The different types of uncertainty pose dissimilar challenges to decision making and hence to the design of impact assessment indicators. As a recent SETAC working group has pointed out, these uncertainties are resolved through value judgments. We list the types of value judgments and explain the choices we have made in our work for EDF's Scorecard project.

524 An Assessment of Environmental Fate and Exposure of Benzene and the Chlorobenzenes in Canada. MacLeod, M. J.* and Mackay, D., The Environmental Modelling Centre, Trent University, Peterborough, ON, Canada. A systematic model of fate and exposure to benzene and the chlorobenzenes which follows a 5-stage process of chemical classification, quantifying of discharge rates and environmental concentrations, evaluative assessment of fate, regional mass balance modelling, and near-field evaluation has been carried out for the southern Ontario region. The EQC model was applied to determine the principal transport and transformation processes experienced by this range of chemicals, which vary considerably in volatility and hydrophobicity. Observed environmental concentrations are in satisfactory agreement with the predictions of the ChemCAN model of chemical fate in Canada. A stochastic, multiple pathway human exposure model which estimates intake of contaminants by residents of the region has been developed and applied to the chlorobenzenes. Results suggest benzene and 1,4-dichlorobenzene are present in the southern Ontario environment at levels sufficient to cause exposures near allowable daily intake levels for the general population of this region. Implications are discussed for evaluation of environmental fate and exposure of other chemical series using the same procedure.

525 **Overall Human Exposure Based on Spatial Range and Persistence.** Bennett, D.H.*, Kastenberg, W.E., McKone, T.E., UC Berkeley, Berkeley, CA. We have developed methods to quantify overall human exposure based on the spatial range and persistence for semi-volatile persistent organic pollutants in a multimedia environment. It is import to know the spatial range and persistence when evaluating the extent of the potential health and ecological impacts of a chemical release. The measure of total human exposure can be used by decision makers in several chemical analyses, including risk assessments, life cycle analyses, and regulatory impact studies. Short or long term effects are determined by the persistence; while the spatial range determines whether a chemical will have a local, regional, or global scale effect. In this research, both persistence and spatial range are determined for a four-compartment steady-state system with air, plants, and two soil layers. The steady-state mass distribution is found to sufficiently represent the actual environment for characterizing persistence and spatial scales, while retaining sufficient simplicity to complete calculations in a tractable form. Persistence depends on the distribution among the different environmental media because decay rates often differ between environmental media. The spatial range is derived analytically from a moving Lagrangian air cell and non-moving compartments. The concentration is reduced with distance based on degradation in air, transfer to and subsequent degradation in vegetation and soil. These methodologies are appropriate for continual, area-source (non-point) emissions, including urban areas with releases from traffic, combustion and industrial activities or regional pesticide use. Using the spatial range, persistence, and mass distribution, a measure of overall human exposure is calculated based on multiple exposure pathways. This approach is limited by the lack of media-specific decay data. Thus, more decay data is needed before models can be made more complex and site-specific.

527 **Measurement and Modeling Implications of Partitioning, Transfer and Transformation Processes at the Plant/Air Interface.** Maddalena, R.L.*, University of California, Davis, CA 95616; T.E. McKone, Lawrence Berkeley National Laboratory and University of California, Berkeley, CA 94720. To understand the role of plants in a multimedia system, the rate of chemical transformation in the plant compartment and the factors controlling the rate and extent of chemical partitioning between the plant and adjacent environmental media need to be measured. An exposure system based on the principle of a continuous stirred tank reactor (CSTR) was designed and used to expose above ground vegetation to semi-volatile air contaminants. Mass balance in the air/plant system is described by an analytical solution of two first-order coupled differential equations representing the time dependent concentrations in each compartment. Measured concentrations of phenanthrene, anthracene, fluoranthene and pyrene in the chamber air and the plant tissue were collected during both the uptake and clearance phase of an exposure experiment and fit to the mass balance by optimizing predicted values for the plant/air partition coefficient, the overall mass transfer coefficient across the plant/air interface and first-order reaction rates in the air and plant compartments. The measured partition coefficients (mol $m^{-3}{}_{[fresh\ plant]}$/mol $m^{-3}{}_{[air]}$) ranged from 5.5×10^5 for phenanthrene to 1.2×10^6 for pyrene. The overall mass transfer coefficient across the air/plant interface (m day^{-1}) ranged from 97 for anthracene to 213 for fluoranthene. In addition to providing information on the partitioning and mass transfer characteristics, the results indicate that a chemical loss pathway may exist at or near the plant/air interface. Implications of this loss pathway are presented and discussed in the context of a multimedia model.

528 **Bioaccumulation of Inorganic Elements by Plants: New Empirical Models.** Efroymson, R. A.*, B. E. Sample, G. W. Suter II, J. J. Beauchamp, M. S. Aplin, and M. E. Will. Oak Ridge National Laboratory, Oak Ridge, TN. Empirical models are required for the estimation of chemical concentrations in plants at contaminated sites where 1) potential risks to herbivores or their predators are of concern and 2) plant tissue concentrations have not been measured. Soil-plant uptake factors that are currently available are not applicable to a wide range of chemical concentrations in a wide range of soils. Regression models were developed to represent the relationship between total concentrations of chemicals in soil and those in above-ground vegetation. The models represent samples from a numerous field sites and soils amended with inorganic salts in the laboratory; most data were obtained from published studies. The chemicals for which models were developed are: arsenic, cadmium, lead, mercury, nickel and selenium. The better-fitting models of concentrations in plants versus concentrations in soil are log-log rather than linear regressions. Multiple regressions with pH as a variable were also developed. In addition, conservative functions for screening assessments were derived. Models were validated using field data collected for ecological risk assessments at two contaminated sites. Plant uptake relationships derived from field studies were compared to those derived from salts amendments. Recommended regressions for use in ecological risk assessments will be presented.

529 **Experimental Studies of Multimedia Model Reliability: Implications of Modeling Error for Human Health Risk Assessment.** Sheldon, A.B.*, Colorado School of Mines, Golden, CO; Dawson, H.E., Colorado School of Mines, Golden, CO; Siegrist, R.L., Colorado School of Mines, Golden, CO. A multimedia lysimeter apparatus was used to measure simultaneous leaching and volatilization over time of toxic volatile organic compounds (VOCs) from soil. Time-average air and leachate chemical concentrations observed during the experiments were compared to time-average concentrations developed using selected intermedia transfer models (ITMs) programmed to simulate the experiments. Several human exposure scenarios were used to calculate cancer risk values for each pair of experimental and modeled time-average VOC exposure concentration values. Human exposure calculations were performed using point estimate input parameters as well as Monte Carlo techniques. ITM reliability was evaluated by considering differences between experiment- versus model-based cancer risk estimates. Differences in point estimates of risk observed for the evaluated soil/environmental conditions and exposure scenarios were large enough to warrant potential concern over model reliability. However, when other sources of risk assessment uncertainty were considered, the magnitude and significance of the observed differences were found to depend on which sources of uncertainty or variability were included in the Monte Carlo simulation. The sensitivity of human health risk predictions to concentration term uncertainty was found to be similar in magnitude to the predictions' sensitivity to human exposure factors and toxicity factors.

530 **Development and Application of a Linked Watershed/Waterbody Model for the Lake Lanier Watershed.** David W. Dilks*, Theodore A. D. Slawecki, Timothy J. Feist, Penelope E. Moskus, R. Scott Wade; LTI, Limno-Tech, Inc., Ann Arbor, MI. Lake Sidney Lanier is threatened by rapid expansion of the Atlanta metropolitan area into its 1000 mi^2 watershed. The objective of this study was to develop a scientifically-based management tool that describes the relationship between future land use activities in the watershed and resulting lake water quality. Specific water quality concerns were lakewide eutrophication impacts and downstream fish toxicity impacts from manganese flux from lake sediments during period of anoxia. The GWLF model was applied to characterize watershed loads, using soils data digitized through a cooperative agreement with the NRCS and land use data derived from satellite imagery. The U.S. Army Corps of Engineers' water quality model CE-QUAL-W2 was modified to include manganese simulation and to consider hydrodynamic branching around mid-lake islands. The models

were calibrated to data from a 1991 Clean Lakes study, and validated to data collected from 1996-1997. A total of 33,000 data observations were available to support model application. The calibrated models were used to evaluate a range of future development scenarios, and have been turned over to the Georgia Department of Natural Resources for further use in water resource management. Study findings include the following: 1) Given a sufficiently robust database, it is possible to describe the water quality processes of concern, as well as to identify specific weaknesses in existing model frameworks; 2) All of the future scenarios being considered for the watershed will result in significant degradation of water quality due to increased runoff from future development; and 3) Intergovernmental co-operation among counties in the watershed regarding future development will be required to protect water quality.

531 **Whole Effluent Toxicity Survey: Assessing the Implementation Status of the Whole Effluent Toxicity Testing Program.** G.M. DeGraeve, G.J. Smith, W.H. Clement and D.O. McIntyre, Great Lakes Environmental Center, Columbus, OH and Traverse City, MI. A Whole Effluent Toxicity (WET) POTW Survey sponsored by the Water Environment Research Foundation was conducted by GLEC to evaluate the implementation status of the WET approach as a water quality protection mechanism within the NPDES program. The objectives were to collect and evaluate regulatory agency and POTW facility data on current WET program practices in the United States, and to identify situations representing effective implementation of the WET program, and circumstances where data indicate a need to improve implementation of the program. WET program management information was first obtained from the regulatory agency representing each state's NPDES permit program, and then by surveying a large national subsample of POTWs required to conduct WET testing. Eight criteria (developed based upon USEPA's TSD and WET Control Policy) were used to assess the implementation status and success of the WET program. Based on the survey responses, it is apparent that the WET program has been implemented throughout the country, and has become an integral component of the NPDES program. The survey results indirectly suggest that the test methods that are routinely used to measure WET are technically sound, and can be routinely performed by competent, well-trained laboratories. However, we found that in certain elements of the WET program, there is meaningful inconsistency in the implementation of the program from jurisdiction to jurisdiction, and there are multiple examples of situations where EPA guidance and policy are not being followed. Areas where implementation of the WET program could be improved in selected regulatory jurisdictions include: the use of "reasonable potential" in determining the need for WET limits; the consideration of the concentration of the effluent in the waterbody after mixing in developing permit limits; the use of enforcement action in the event that there is a pattern of WET violations; and the use of professional judgement in addressing WET issues.

532 **Defining the Relationship Between Whole Effluent Toxicity Testing and Instream Toxiciy.** Gerardi, C.*, Tetra Tech, Inc., Owings Mills, MD; Diamond, J., Tetra Tech, Inc., Owings Mills, MD. A database was compiled consisting of over 1300 whole effluent toxicity (WET) tests and 270 benthic macroinvertebrate assessments conducted for 250 facilities across the U.S. These data were subjected to various retrospective statistical analyses to define relationships between WET results and actual instream biological condition. Results indicate that the percentage of failed tests (using standard pass/fail criteria), for all freshwater WET endpoints, were directly related to the number of tests performed. Instream impairment was related to WET results if: (a) the effluent comprised >60% of the 7Q10 stream flow; (b) was associated with fair or better instream habitat quality (c) conducted >3 WET tests, and (d) had failed at least 25% of their WET tests. Fish NOEC endpoints, and *C. dubia* acute and chronic endpoints, were not effective predictors of instream conditions. Fish LOEC/average effluent concentration (IWC), or acute percent mortality in undiluted effluent, were better predictors of instream conditions. An average chronic NOEC/IWC (for both species averaged for each facility) was significantly related to the biological condition instream for effluent dominated sites. Multi-year data suggested a time lag between WET results and instream effects depending on the type of effluent pollutants discharged, pollutant bioavailability, and instream dilution. The use of an NOEC at design IWC for chronic test compliance appeared to be overly stringent in most cases. The LOEC and design IWC or the NOEC and the IWC during the time of testing appeared to be more predictive of instream effects. In order to more accurately portray pollutant bioavailability instream, laboratory tests should be accompanied by some form of *in-situ*, ambient toxicity monitoring, or biological assessments.

533 **Development of Whole Effluent Toxicity Tests with New Zealand Native Species.** Hall, J.A.*, NIWA, Hamilton, New Zealand, Nipper, M.G., Texas A&M University-Corpus Christi, Corpus Christi, Texas, USA. Whole Effluent Toxicity (WET) tests have become a requirement for discharge consents by regulatory bodies in New Zealand. This has highlighted the need for toxicity test protocols with New Zealand species of marine and freshwater algae, invertebrates and vertebrates. Suitable protocols existed for native marine invertebrates (echinoid embryo development test) and freshwater algae (*Selenastrum capricornutum* growth test). This study focused on the identification of appropriate species and development of protocols for marine algae and fish, and freshwater invertebrates and fish. In each group a minimum of four species were screened for sensitivity with two inorganic (Cd and Zn) and two organic (phenol and SDS) reference toxicants. Each species was also evaluated for ease of handling and for year-round availability, either by field collection or feasibility of laboratory culturing. The species selected were the freshwater and marine fish, *Gobiomorphus cotidianus* and *Rhombosolea plebia*, respectively, the marine micro-alga *Dunaliella tretiolecta*, and two species of freshwater invertebrates, the cladoceran *Ceriodaphnia dubia* and the amphipod *Paracalliope fluiviatilis*, representative of still and running waters, respectively. Bimonthly testing was conducted during one year with the reference toxicants Zn and SDS, to establish any seasonal trends in sensitivity for the field collected species, and to compare the sensitivity of the different test methods. No seasonal trends have been observed. The most sensitive marine methods were the micro-alga, for Zn, and the fish for SDS. Among the freshwater methods the cladoceran were the most sensitive to both Zn and SDS. This emphasises the importance of conducting a variety of WET tests with native species of different trophic levels, for use in regulatory discharge consents.

534 **Aquatic Ecological Risk Assessment of Episodic Sewer Overflows in Sydney, Australia.** Toll, J.*, MacLellan, D., Stivers, C., Keithly, J., Kluck, M. and Logan, L., Parametrix, Inc., and Bickford, G. and Hansen, J., Sydney Water Corporation. This paper discusses technical issues associated with assessing aquatic ecological risks from spatially and temporally distributed episodic events, on a regional scale, against a backdrop of urban anthropogenic impacts. An aquatic ecological risk assessment of sewage overflows in greater Sydney, Australia is presented as a case study. The assessment identified copper; silver; zinc; 2,4-dichlorophenol; α-BHC; β-BHC; chlordane; chlorpyrifos; diazinon; dieldrin; hexachlorobenzene; ammonia; nitrite; hydrogen sulfide; total suspended solids and dissolved oxygen as stressors of concern. Loads of copper, zinc, and pesticides are derived primarily from stormwater. Ammonia, nitrite, and silver are associated with both sewage overflows and stormwater. Abatement of sewage overflows would likely have some benefit by reducing the risk from these chemicals. Dissolved oxygen and total suspended sediments concentrations were found to present some risk to aquatic life at some of the receiving water sites examined. Benthic survey and bioassay results corroborated the risk assessment's predictions. The benthic survey found communities generally depauperate. Sewage overflow sites and urban reference sites were similarly impacted. Bioassays showed that receiving waters upstream of sewage overflows were toxic in wet weather, and at some locations in dry weather. Sediment bioassays indicated toxicity generally is not expected for organisms whose sensitivities are similar to amphipods.

535 the Role of Pulp Bleaching in MFO Induction in Rainbow Trout. Coakley, J. D.*, Queen's University, Kingston, ON, Canada; Hodson, P. V., Queen's University, Kingston, ON, Canada; van Heiningen, A. R. P., University of New Brunswick, Fredericton, NB, Canada; Cross, T., Queen's University, Kingston, ON, Canada. Fully bleached kraft pulp mills produce effluents that cause induction of hepatic mixed function oxygenase enzymes (MFO) in rainbow trout. Using bioassays of MFO induction in trout, spent filtrates from pulp bleaching were assessed for their MFO induction potency. Filtrates were collected from kraft mills in Cornwall, Ontario and Smooth Rock Falls, Ontario to assess the potency of filtrates from hardwood and softwood bleaching, respectively. Mill-bleaching filtrates induced MFO activity at concentrations greater than 5.6% v/v filtrate, and filtrates from softwood pulp bleaching appeared more potent than filtrates from hardwood bleaching. In laboratory bench-scale bleaching experiments, pulp from a kraft mill in Miramichi, New Brunswick was bleached via an industry-standard 5-stage chlorine dioxide bleaching sequence. The filtrates were collected and used in fish bioassays to assess MFO-inducing potency. Potency of laboratory filtrates to induce MFO activity varied depending on the bleaching stage, with the first alkaline extraction stage having the highest potency.

536 Organophosphate Pesticide Removal in Wastewater Treatment. Drury, D. and Baker, C., Chino Basin Municipal Water District, Fontana, CA; Moore, T., Risk Science, Brentwood, TN. Chino Basin Municipal Water District experienced toxicity at its Regional Plant No.2 (RP-2). Using *Ceriodaphnia dubia* as the test organism, twenty-four acute toxicity events occurred between April and November 1996. To reduce the toxicity, Central Avenue trunkline flows were diverted from RP-2 to the Carbon Canyon Water Reclamation Facility (CCWRF) which had no toxic events in 1996. Treatment at CCWRF was thought to be possible because of its conservative design, which allowed for the activated sludge process to be operated with long SRTs (>40 days) and hydraulic detention times (>3 hours actual) and high MLSS (>3300 mg/L). It was hypothesized that the long SRTs and high MLSS would result in the maximum biosorption and biodegradation of organophosphate pesticides, such as chlorpyrifos and diazinon. To measure the effectiveness of the activated sludge in removing chlorpyrifos and diazinon, the influent and effluent of CCWRF were analyzed three times per week beginning March 1997. The concentrations of chlorpyrifos entering the plant were as high as 2.3 ug/L. The effluent chlorpyrifos concentrations averaged 0.048 ug/L. Occasionally, the effluent chlorpyrifos concentrations were high enough to cause toxicity. For diazinon, the influent concentrations were as high as 12.2 ug/L and the effluent averaged 0.033 ug/L. CCWRF was able to achieve >95 percent removal of both chlorpyrifos and diazinon. During the same time period in 1997 there were ten incidents of acute toxicity. Virtually all of the chlorpyrifos removal occurs in the activated sludge. For diazinon, approximately one-third of the removal occurs in the activated sludge. The remaining two-thirds occurs in chlorination. The pesticide removals were not high enough to completely remove toxicity.

537 Assessment of Toxicity from Municipal Effluents Discharged into the Hawksbury Nepean River, New South Wales, Australia, Using *Ceriodaphnia dubia* and *Selenastrum capricornutum*. H. Bailey and J. Elphick, EVS Environment Consultants, North Vancouver, BC, R. Krassoi, Sinclair, Knigh and Merz, St. Leonards, NSW, A. Lovell and G. Bickford, Sydney Water Corporation, Sydney, Australia. Samples of effluent discharged from 18 sewage treatment plants in the Hawksbury-Nepean river were evaluated for acute and chronic toxicity using *Ceriodaphnia dubia* and *Selenastrum capricornutum*. Toxicants identified using TIE procedures included the pesticides diazinon, chlorfenvinphos and chlorpyrifos. Ammonia also contributed to toxicity at one site. The contaminant(s) responsible for transient toxicity to C. dubia at four sites were not identified, although surfactants, including MBAS, CTAS and sludge dewatering agents, were ruled out. Only two samples exhibited toxicity to S. capricornutum the cause(s) were not identified other than they were non-polar organic constituents that dissipated over time. Further testing is underway to confirm these results and to determine the identities of the transient toxicants.

538 Toxicity of Total Dissolved Solids (TDS) From Mine Effluent Related to Water Quality Criteria. Chapman, P. M.*, H. Bailey and E. Canaria., EVS Consultants, N. Vancouver, B.C., Canada. Measurement of total dissolved solids (TDS) represents an integrative measure of the concentrations of common ions (e.g., sodium, potassium, calcium, magnesium, chloride, sulfate and bicarbonate) in freshwaters. Toxicity related to these ions is due to the specific combination and concentration of ions, and is not predictable from TDS concentrations. However, despite this, regulatory bodies have set TDS limits for freshwater bodies, generally related to chloride and other salt concentrations in a stream. Most TDS toxicity studies have been conducted with the aquatic white rat organisms used routinely for effluent toxicity tests (e.g., daphnids, fatheads). There is little information on TDS toxicity to other organisms. We conducted chronic toxicity tests with more environmentally relevant organisms (larval chironomids and eyed trout eggs) to assess their TDS tolerance relative to effluent from two Alaskan mines. Both effluents are characterized by high TDS content in addition to trace metals, however one effluent had much higher alkalinity than the other and contained a higher relative concentration of chloride, whereas the other was primarily sulfate-dominated. Tests with synthetic effluent demonstrated non-toxicity in the former case at >2,000 mg/L TDS to both test organisms; the latter effluent was found to be similarly non-toxic to developing trout eggs but chironomids showed effects above 1,000 mg/L TDS. These two tests, together with information on the health of field populations (fish and benthic invertebrates), appear to be useful and relevant for determining site-specific whole effluent TDS concentrations.

539 Toxicity Testing Considerations for Low Ionic Strength Effluents. Hartzell, L., Goodfellow, W.*, Elseroad, J. EA Engineering, Science and Technology, Inc., Sparks, Maryland. Whole effluent Toxicity (WET) testing has become a routine part of NPDES permit compliance in the United States. For most instances, effluent can be easily evaluated for acute toxicity using the standard protocols without much consideration as to the effluent's characteristics. However, as WET procedures are used for evaluating effluents for chronic toxicity that are marginally toxic (e.g., only effect growth or reproduction), greater attention to test procedures as well as effluent characteristics are necessary. One aspect of WET programs that exists is what to do when an effluent is discharged to an effluent dominated system (e.g., high IWC) and the effluent is of extremely low ionic strength. These type of discharges are common with condensate type discharges or in treatment systems that use natural or synthetic exchange resins. This paper presents toxicity testing considerations and procedures for low ionic strength effluents. Through the use of case studies, recommendation as to technical and regulatory considerations will be provided.

540 Toxicity of a Petroluem Refinery Wastewater to an Estuarine Shrimp, *Mysidopsis bahia*. Johnson, G.M.*, Shell Martinez Refining Company, Martinez, CA; Glaze, D.E. Shell Martinez Refining Company, Martinez, CA; Van Compernolle, R. Shell Development Company, Houston, TX. This research investigated whether or not the acute toxicity to an estuarine shrimp, *Mysidopsis bahia* (mysid), upon exposure to treated refinery wastewater was caused by a group of organic acids called naphthenic acids. Previous studies by this discharger using threespined stickleback and rainbow trout showed that naphthenic acids were the primary contributor to toxicity in this wastewater. This research also investigated the possibility of using supplemental granular activated carbon (GAC) to reduce observed levels of acute toxicity to mysids. This research relied on a combination of evaluating known parameters measured in the treated wastewater for toxicity to mysids and performing a series of experiments designed to confirm the presence of naphthenic acids toxicity to mysids. These experiments included: 1) determining the naphthenic acids mysid LC50 concentration, 2) evaluating naphthenic acids toxicity dependence on pH, 3) performing screening tests to detect ion imbalances in the treated wastewater, 4) evaluating the full-scale GAC units' effectiveness at reducing toxicity to mysids, and 5) determining if supplemental GAC treatment can further reduce toxicity to mysids. This study showed that the mysid toxicity is primarily caused by naphthenic acids present in the treated wastewater and that supplemental GAC treatment may be used to further reduce toxicity. This study also showed that ion imbalances may be a minor contributor to mysid toxicity.

541 **Field Evidence for Linking Altosid Applications with Increased Deformities in Southern Leopard Frogs.** Sparling, D. W.*, USGS Patuxent Wildlife Research Center. The objective of this study was to determine if Altosid, a formulation of 4% methoprene used for mosquito control, could be a cause for the types of amphibian deformities occurring in parts of North America. The formulation was repeatedly sprayed on six constructed macrocosms during late spring and summer following label directions. Six additional macrocosms were sprayed (also at label specifications) with Abate-4E, containing the organophosphate pesticide temephos, and six were sprayed with water (controls). In early September juvenile frogs and metamorphing tadpoles were collected and examined for deformities. In all, 91 juvenile and metamorph southern leopard frogs (*Rana utric2ularia*) were collected from Altosid sprayed wetlands with 14 (15%) demonstrating deformities. Seventy-seven juveniles and metamorphs were collected from control wetlands with three (4%) showing deformities. Only six juveniles and metamorphs were collected from Abate-4E wetlands and none showed deformities. Deformities included missing or deformed hind limbs (9 of 10 involving only the right hind limb), missing eyes, and abnormal color. The differences in rate of deformities was dependent on treatment (X^2=6.44, $p < 0.02$). The number of leopard frogs caught per unit effort (tadpoles and juveniles) differed among treatments (p=0.032) with Abate-4E wetlands producing fewer individuals per capture effort than either Altosid or control wetlands. Results from the summer of 1998 which included an experiment to determine when malformations occur during development are reported.

542 **Effects of Ultraviolet B Radiation and Ambient Metals on Native Anuran Development.** Hansen, L.J., University of California, Davis. Amphibian decline is both a global and regional issue. Many areas worldwide, including western North America, have chronicled an incidence of amphibian decline, often in numerous genera. Many theories have been proposed as to the potential causes of amphibian declines but no definitive conclusions have yet been drawn. It is most likely that there is no universal agent and it is also most probable that combinations of factors are responsible for even regional extirpations. One theory proposed has been that the increase of Ultraviolet B (UVB) radiation as caused by stratospheric ozone depletion is negatively impacting developing anuran embryos. However the majority of the causative work in this area, while under ambient lighting, has been done in the absence of dosimetry. Additionally the effects of UVB with other stresses has also not been well explored. In order to determine the interactive effects of quantifiable UVB radiation and metals on developing native anurans (Pacific Treefrog, *Hyla regilla*) a series of experiments were conducted to compare not only these effects but local population adaptations to these stressors as well. High and low elevation populations of amphibians were exposed to four treatment levels of UVB (subambient, low ambient, high ambient and enhanced) and three treatment levels of metals (non-detect, ambient and enhanced) within a full-spectrum solar simulator. Exposures began prior to gastrulation and ended at Gosner Stage 28, with partial formation of limb buds, front and back. Endpoints include growth, development, hatching success, pigment induction, histopathology and molecular genetic identification. Exposures to UVB show local population adaptations with low elevation population tadpoles being more sensitive to UVB at ambient levels as indicated by slowed development and histopathological and pigment induction abnormalities. Sublethal endpoints are particularly useful in this species which inhabits ephemeral ponds, as failure to reach metamorphosis prior to pond dry down removes that cohort from the population thereby decreasing reproductive recruitment in subsequent years.

543 **Effects of Methoprene and Ultraviolet Light on Survival and Development of *Rana pipens*.** Ankley, G.T.*, Tietge, J.E., DeFoe, D.L., Jensen, K.M., Holcombe,G.W., Durhan, E.J., Diamond,S.A., U.S. EPA, Duluth, MN. There recently have been increasing observations in North America of a suite of relatively specific hindlimb deformities in several anuran species. These deformities include supernumerary limbs and missing limbs, limb segments, or digits. The objective of this study was to assess two stressors hypothesized as responsible for limb malformations in amphibians: methoprene, an insect growth regulator which, through interaction with the retinoic acid signaling system, could possibly cause limb deformities, and ultraviolet (UV) light. Northern leopard frogs (*Rana pipiens*) were exposed to several different concentrations of methoprene both in the absence and presence of UV light designed to mimic the (UV) wavelength spectrum present in sunlight. At the highest methoprene concentration tested, both in the absence and presence of UV light, severe developmental effects were observed, with all organisms dying within 12 to 16 d of test initiation. Irrespective of methoprene treatment, a very high percentage (ca., 50%) of animals held under the UV light for ≥24 d developed hindlimb deformities. These deformities usually were bilateral and sometimes completely symmetrical, and consisted of missing limb segments and missing or reduced digits. The developmental period of greatest sensitivity to the UV light occurred during very early limb bud development. The significance of these findings in terms of deformed frogs in the field is uncertain. For example, while the deformity types observed (i.e., missing limb segments/digits) are similar to those seen in some field specimens, the UV light treatment did not cause the full range of observed malformations in animals from the field. Despite these uncertainties, our findings suggest that UV light should be further considered as a plausible factor contributing to amphibian deformities in field settings.

544 **Application of the FETAX Protocol to Surface Waters Associated with Malformations in Native Anuran Populations.** Tietge JE*, Ankley GT, Holcombe GW, DeFoe DL, Jensen KM, U.S. EPA, Duluth, MN. Surface water from a site in Minnesota with malformed anuran populations was tested for developmental effects with the Frog Embryo Teratogenesis Assay: *Xenopus* (FETAX). The purpose of this work was to determine if developmental effects could be used as part of an effects-based fractionation approach to isolate and identify possible developmental toxicants in chemically complex surface waters. Under standard FETAX conditions, 25 organisms are placed in a 10 mL volume of test solution which is renewed daily. At 96 h, the organisms are assessed for abnormal development. Under these conditions, the site water effected cranio-facial and gut development and resulted in generally retarded growth. However, all of the effects were eliminated by several different treatments, including: testing the organisms in a larger volume (100 mL) of site water, adding salts to the site water that are used to make the standard FETAX testing solution, evaporation of site water to increase the concentration of dissolved solids, and by dilution of the site water with FETAX testing solution. These results are consistent with the hypothesis that the low ion concentrations found in this particular site water are responsible for developmental effects observed in the FETAX assay. This hypothesis was strengthened further when a reconstituted site water made with deionized water and reagent grade salts resulted in the same effects. Taken together these data suggest that the recommended protocol for FETAX does not deal adequately with the inherent variability of field collected waters and that more care should be taken when using such waters. The toxicity observed in these FETAX studies are probably not relevant to the malformations observed in native anuran species.

545 **Water and Sediment from Selected Sites in the Rio Grande River Induce Anencephaly in Frog Embryos.** Propst, T.L., Fort, D.J., Schetter, T.A., and Stover, E.L., The Stover Group, Stillwater, OK; Bantle, J.A. and Dumont, J.N., Oklahoma State University, Stillwater, OK. Water and sediment samples from three sites in the Rio Grande River were collected to evaluate potential effects on amphibian development and maturation using a modified Frog Embryo Teratogenesis Assay - *Xenopus* (FETAX) protocol. Site 1 was located immediately downstream from Brownsville, Texas/Matamoros, Mexico. Site 2 was located nearly 25 miles

upstream and between Sites 1 and 3. Site 3 was located immediately downstream from Reynosa, Mexico. Results of testing indicated both Sites 1 and 3 induced developmental toxicity. Water samples from Site 1 induced mortality and malformation rates of 52% and 100%, respectively. Whole sediment tests of Site 1 samples produced complete embryolethality (anencepahlic at time of death). Abnormalities induced by exposure to Site 1 water included miscoiling of the gut, kinking of the notochord, mal-development of the eye, and anencepahaly. Interestingly, high rates of anencephaly have been reported in infants born in the Brownsville/Matamoros area during the late 1980s and early 1990s. Water samples from Site 3 induced mortality and malformation rates of 8.3% and 14.6%, respectively. Sediment samples from Site 3 induced mortality and malformation rates of 30.0% and 100%, respectively. Abnormalities induced by Site 3 samples included abnormal gut development, mal-development of the craniofacial region, visceral edema, and mal-development of the heart. Samples collected from Site 2 did not induce developmental toxicity. Overall, these studies indicated that water and sediment from the Rio Grande River are capable of inducing abnormal development in *Xenopus* and that specific hot spots within the river probably exist.

546 **FETAX Analysis of Contaminated River Water and Sediments.** Marquez-Bravo, L. G.*, Dumont, J. N.** and Bantle, J. A.**, *Instituto Mexicano de Technologia del Agua, Jiutepec, Mexico and **Department of Zoology, Oklahoma State University, Stillwater, OK 74075. Water and sediment samples were taken from suspected contaminated areas along the Rio Grande river. The sediment samples were extracted with either acetone or dimethylsulfoxide, diluted to a final concentration of 1% solvent and tested with FETAX (Frog Embryo Teratogenesis Assay: *Xenopus*) that measures the endpoints of embryolethality, malformations, and embryo growth. FETAX was conducted with and without the addition of a Metabolic Activating System (MAS) that consists of induced rat liver microsomes and appropriate enzymes. MAS is designed to mimic mammalian metabolic activity. Results indicate that embryolethality is not substantially increased over controls in the non-MAS tests but about 15% of the samples show increased embryolethality in tests which included MAS. On the other hand, malformations are increased in about 45 % of the samples in non-MAS tests. The malformation rate increases in 75% of the samples when MAS components are added. Typical malformations include severe abnormalities in eye, edema, and abnormal development of the head, face and gut. Growth, i.e., length attained by the embryos, is significantly decreased in 70% of the samples tested without MAS and in 93% of the samples to which MAS components have been added. These results suggest that areas along the course of the river are significantly contaminated and that the addition of MAS components to mimic mammalian metabolic activity increases both the embryolethality and teratogenicity.

547 **Morphological and Biochemical Effects of Environmental Estrogens on Frog (*Xenopus laevis*) Metamorphosis.** Cheek, A. Oliver*, Rider, C.V., Holstein, M.A., McLachlan, J.A., Tulane University Center for Bioenvironmental Research, New Orleans, LA. Environmental estrogens are capable of disrupting reproductive physiology in amphibians. Recently, the endogenous estrogen, estradiol-17§ (E2), has been shown to delay frog metamorphosis. Since frog metamorphosis is primarily controlled by thyroid hormones, we used metamorphosis as a model for environmental estrogen interaction with the thyroid axis. We exposed premetamorphic tadpoles of the African clawed frog, *Xenopus laevis*, to steroidal and environmental estrogens and measured growth, rate of metamorphosis, and thyroid hormone receptor (TR) expression. Thyroid hormone auto-induces TR expression, so alterations in TR concentration indicate interaction with the thyroid axis. Steroidal estrogens consistently delayed spontaneous and thyroid hormone-induced metamorphosis when given during early premetamorphosis. Interestingly, E2 treatment enhanced TR expression in animals treated with thyroid hormones (triiodothyronine (T_3) and thyroxine (T_4)), but decreased TR expression in animals undergoing spontaneous metamorphosis. o,p'DDD, a metabolite of DDT, inhibited thyroid-hormone induced metamorphosis only at high (1 μM) concentrations. 3,3',4,4'tetrachlorobiphenyl (PCB 77) tended to delay T_4-induced metamorphosis and decreased T_4-induced expression of TR. PCB 77 did not alter metamorphic rate or TR expression in tadpoles undergoing spontaneous metamorphosis. Delayed metamorphosis is correlated with altered TR expression, indicating that steroidal and environmental estrogens interact with the thyroid axis.

548 **Pond Water Samples from Minnesota and Vermont Disturb Limb Development and Thyroid Function (Tail Resorption) in *Xenopus*.** Fort, D.J.*, Propst, T.L., Schetter, T., and Stover, E.L., The Stover Group, Stillwater, OK; and Burkhart, J.G., NIEHS, Research Triangle Park, NC. Four test sites in Minnesota and three sites in Vermont were selected to evaluate effects of pond water on limb development and thyroid function in *Xenopus laevis*. Hind limb development was monitored during a 30-d (stage 8-54) static renewal exposure regime and tail resorption was monitored during a 14-d exposure regime from day 50 to day 64 (stage 60-64) using video image capture. Two of the four test sites in Minnesota induced abnormal limb development characterized by atrophy of the soft tissue and reduction deficits, primarily distal to the femur. The fourth test site in Minnesota which did not induce abnormal limb development did cause an appreciable developmental delay. In addition, three of the four test sites in Minnesota induced a significant slowing of the tail resorption rate. In each case, co-administration of thyroxin with the test site sample reversed the slow response to rates comparable to the reference site. One of the three sites in Vermont induced abnormal limb development which was characterized by atrophy of the soft tissue and random skeletal defects. Each of the three sites showed dramatically slowed limb development compared to the paired reference site. Each of the three test sites from Vermont induced significantly slowed tail resorption rates compared to the paired reference site. In the case of three of the four test sites, co-administration of thyroxin reversed tail resorption inhibition increasing the rates to near that of the paired reference sites. Results indicated that longer-term effects on development and maturation of amphibians can be monitored in the laboratory using *Xenopus*.

549 **Evidence for Potentiation among Environmental Factors That Contribute to Malformations in Frogs.** Burkhart, J.G.*, Gallagher, K., NIEHS Research Triangle Park, NC; Fort, D.J., Propst T.L., The Stover Group, Stillwater OK; Helgen, J.C. MPCA, St. Paul, MN. Investigation of water samples from reference sites and sites associated with the increased incidences of malformations in frogs has revealed that factors from sites with low rates of malformation are capable of potentiating the toxicity of water from affected sites when assayed in *Xenopus*. C-18/methanol elution of water from affected sites has produced fractions that are toxic and malforming to *Xenopus* embryos. Compounds have been identified within those fractions. Similar treatment of water from certain reference sites does not produce toxic fractions. However, there are nontoxic fractions from the reference sites that potentiate the incidence and severity of deformities produced by unprocessed water and methanol fractions from the affected sites. Compounds have been identified in the potentiating fractions and there is little similarity with chemicals identified in the affected sites. The primary composition of the potentiating fractions appears to be natural compounds that resemble sterols and phytoestrogen-like structures. One of the major questions to come from this line of investigation is whether or not levels of intrinsic compounds such as natural estrogens or microbial autoregulators may play a role in the outcome of a subsequent exposure for a particular site or region.

550 **Characterization of the Causes of Abnormal Development of Frog Embryos in Pond Water and Sediment from the State of Minnesota.** Fort, D.J., Propst, T.L., Schetter, T., Stover, E.L., The Stover Group, Stillwater, OK; Burkhart, J.G., NIEHS, Research Triangle Park, NC. In an effort to elucidate the causes of abnormal development in frog embryos exposed to water and sediment collected from a pond in which significant numbers of abnormal frog specimen were

identified, toxicity characterization studies were performed. Toxicity characterization included physical/chemical fractionation of the samples similar to that used in conventional Toxicity Identification Evaluations (TIE) studies using FETAX, matrix evaluation, and chemical-specific analyses of potential toxicants in the samples. Fractionation processes included pH adjustment, filtration, aeration, solid phase extraction (C18, ion exchange, and zeolite), and thiosulfate chemical addition. Characterization found that the C18 and the mixed-bed ion exchange solid phase treatment were capable of removing ca. 50% and 45% of the toxicity observed. Filtration reduced toxicity slightly. Methanol fractionation of toxicants loaded on the C18 column were eluted using a step gradient ranging from 25% methanol to 100% methanol. FETAX studies concluded that the toxicants were bimodally fractionated into the 80 and 85%, and the 100% methanol fractions. GC-MS analyses identified a series suspected toxicants in the combined 80 and 85% fractions and 100% fraction. Toxic levels of nickel were detected and confirmed with FETAX. Although it was not the direct cause of toxicity, the matrix appeared to play a critical role in exacerbating the toxicity observed based simulation studies. These studies indicated that a chemical-specific caused of abnormal frog development exists in pond studied and that the matrix of the pond water likely plays a significant role in the behavior of the toxicants.

551 **Natural and Anthropogenic Chemical Fluxes in Upper Savannah River System Reservoirs.** Elzerman, A. W., Fjeld, R. A., Coates, J. T., Environmental Engineering and Science, Clemson University, Clemson, SC, 29634-0919. The system of reservoirs constructed on the upper Savannah River offer a valuable study system for sources and fates of both natural and anthropogenic chemicals. Since before 1980 we have investigated major ions and acid rain components in relation to weathering rates and observed changes in the series of reservoirs from the mountain region on down the progression through the piedmont region. We have also compared these major ion chemistries to other regional lake systems. The fourth reservoir in the chain, Lake Hartwell, has also received inputs of specific anthropogenic chemicals (PCBs and various radionuclides) which are of interest for their potential environmental and social effects as well as for investigation of fate and effects and as tracers of reservoir processes. Mercury investigations indicate the importance of both natural and anthropogenic effects.

552 **Pollution History of the Savannah River Estuary.** Alexander, C., Ertel, J., Lee, R., Loganathan, B., Smith, R., Wakeham, S., Windom, H. (all at Skidaway Institute of Oceanography, Savannah, GA 31411). As part of the NOAA National Status and Trends Program, 13 cores were collected from the Savannah River Estuary for the production of historical pollutant profiles. These cores, representing intertidal salt marsh, subtidal channel and abandoned boat-slip environments, were dated using Pb-210 geochronologies and analyzed for the metals Al, Ag, As, Cd, Cr, Cu, Fe, Hg, Mn, Ni, Pb, Sb, Se, Sn, and Zn, as well as pesticides, polychlorinated biphenyls, polycyclic aromatic hydrocarbons, and butyltins. Four contaminant-time patterns were identified. For Ag, Cd, and Zn, profiles show an increase in concentration up to the present time, indicating an increasing loading from non-point-source pollution. Cr and DDT isomers exhibit distributions characterized by maxima that get progressively shallower (and younger) down-estuary, representing redistribution of contaminated material. In one core, Hg exhibits a sharp subsurface maxima, indicating a localized anthropogenic input that has since been controlled. Pb, PCBs and PAHs exhibit subsurface maxima representing peak inputs prior to the institution of environmental regulatory controls. In comparison to densely populated and industrialized regions, the concentrations of most anthropogenic chemicals found in cores from the Savannah Estuary are low. Decreases in these components over the past few decades suggest that pollution control regulations have been effective, even while industrial and population growth has occurred. However, levels of inorganic pollutants are approximately twice what they were in the previous century and what they are in contemporary nearby settings.

553 **A Proposed Stream Water Quality Study in the Savannah River Basin.** Smith. C.*, Pope J., Smith P., Brockway D., US EPA, Athens GA; Payne W. and Kollig H., SEEP, US EPA, Athens, GA. There is concern that surface water streams in the Savannah River Basin are contaminated with various pollutants. A proposed water quality characterization study is being planned for major tributaries in the Savannah River Basin for use in model development and testing and for development of Total Maximum Daily Load (TMDL) protocols. The Savannah River Basin was selected by EPA's National Exposure Research Laboratory's Ecosystems Research Division in Athens, Georgia, as the Near Laboratory Ecological Research Areas (NLERA). Major stressors in the basin include agricultural operations and forest production (nutrients, pesticides, animal waste, sediment, bacteria); municipal and industrial waste water discharges (sewage, chemical, textile, paper, etc.); landform modifications related to water impoundments (water storage, power generation); and military bases. The Savannah River Basin is a 10,577 square mile area located along the border of three states with 5821square miles in Georgia, 4581 square miles in South Carolina and 175 square miles in North Carolina. The Savannah River originates in the mountains of Georgia, South Carolina and North Carolina and flows south-southeasterly about 300 miles to the Atlantic Ocean near the port city of Savannah GA. In its upper mountain area are several pristine lakes for power generation (Lake Burton, Tallulah Falls, and Yonah) and in the mid-reaches, the flow of the river is regulated by numerous reservoirs, including three large, multi-purpose US Army Corps of Engineers reservoirs (Hartwell Lake, Richard B. Russell Lake, and Strom Thurmond Reservoir) and two large private power reservoirs in the upper-reaches (Lake Keowee and Lake Jocassee). Numerous farm ponds are located throughout the basin. Approximately 45 miles of the lower Savannah River are influenced by tidal action. Results of the proposed study will be provided.

554 **South Carolina's Watershed Water Quality Management in the Savannah River Basin.** Lackey, A. E., SC DHEC, Columbia, SC. In 1991, the SC DHEC Bureau of Water implemented the Watershed Water Quality Management Program (WWQMP) in order to more efficiently protect and improve the quality of South Carolina's surface waters. The WWQMP organizes water quality management activities in each of the state's eight major river basins on a five-year cycle, which includes intensive monitoring, public involvement, water quality assessment, wasteload allocation, development of watershed strategies, permitting, and implementation. The Savannah River Basin is currently in its second watershed management cycle. This basin covers approximately 10,577 square miles, including portions of Georgia, North Carolina, and South Carolina, and passes through four physiographic provinces (Blue Ridge Mountains, Piedmont, and Upper and Lower Coastal Plains). SC DHEC maintains an extensive monitoring network in the 4,581 square mile South Carolina portion of the basin, collecting water quality samples at over 100 stations and assessing the health of macroinvertebrate communities at close to 70 sites. These data are used to identify waters that do not meet water quality standards and for which SC DHEC will develop total maximum daily loads (TMDLs) to address causes of impairment. South Carolina's 1998 list of impaired waters includes approximately 50 sites in the Savannah River basin that are not expected to meet water quality standards after implementation of currently required controls. The majority of these waters are impaired due to elevated levels of fecal coliform bacteria, although copper and zinc are also causes of impairment in several waters. An overview of the state's water quality management efforts, the quality of waters in the Savannah River basin, and plans for TMDL development will be given.

555 **The CRESP Experiment: Implications to Ecological Research.** Goldstein, B.D.*, Powers, C., McGrath, L.F., Environmental and Occupational Health Sciences Institute (EOHSI)**, Piscataway, NJ; Karr, J.R., University of Washington; Burger, J., Rutgers University. The Consortium for Risk Evaluation with Stakeholder Participation (CRESP) is an experiment in performing stakeholder-related, risk-based research to respond to environmental challenges at DOE sites.

CRESP is located primarily at two academic environmental health sciences programs at EOHSI and University of Washington. It is organized in eight task groups, one of which is ecological health. James Karr heads the Environmental Health Task Group at the University of Washington and Joanna Burger at EOHSI. Their exemplary work includes extensions of Karr's Index of Biological Integrity to terrestrial systems at Hanford and its application to aquatic systems at Savannah River. The approach used helps define ecological dose-response curves which help understand the alteration of systems by various human activities. The work of the Ecological Health Task Groups are particularly notable for their collaboration with the SREL and PNNL, government agencies, tribal and citizen's groups and their interactions with other task groups. The extent of these interactions is highlighted by the work of Joanna Burger in the understanding of fishing behavior and consumption patterns, methods of cooking and potential hazards from consuming fish from the Savannah River. These studies differ from traditional ecological research in their ability to integrate health effects, exposure assessment and survey research to address both research and policy questions. This leads to more comprehensive research that directly impacts policy. Because of the interactive nature of CRESP research, there has been an ability for ecological research to more rapidly and directly impact regulatory guidance. **EOHSI is a joint program of UMDNJ-Robert Wood Johnson Medical School and Rutgers University. Work supported by CRESP through the Department of Energy, AI#DE-FC01-95EW55084

556 **Biological Data Collection for Ecological Risk Assessment.** Dyer, S. A.* and Paller, M. H. Paller, Westinghouse Savannah River Company, Aiken, SC; M. Roy, Radian Corporation, Austin TX; J. J. Nelsen, Department of Energy, Aiken, SC. The Savannah River, five major stream systems of the Savannah River Site (SRS), and the Savannah River Swamp constitute the Integrator Operable Units (IOUs) for the SRS Environmental Restoration Program. The IOU evaluation requires development of a Screening-Level Ecological Risk Assessment (SERA). The purpose of the SERA is to identify constituents that may pose a risk to ecological receptors and focus subsequent investigations. The SERA is a three tiered weight-of-evidence evaluation consisting of: (I) a comparison of media concentrations to ecological screening benchmark values; (II) background screening for surface water, sediment, and fish tissue constituents; and (III) biological data evaluation including fish assemblage data, fish necropsy data, and other available biological data. SERA results for lower Steel Creek showed the potential for adverse effects due to hazard quotients of 1.5 for copper and 5.8 for zinc in surface water. Mercury (at 1.63 mg./kg), cesium-137 (at 2.29pCi/g), and tritium (at 0.838pCi/g) were identified as fish tissue constituents of concern. Fish index of biotic integrity (mean of 38) and health index necropsy data indicated that lower Steel Creek was similar to the reference area. The conclusions drawn were that further media and biological data are required due to the media screening evaluation and the preliminary fish tissue analyses that confirm the presence of contaminants in fish tissue. Effects may be occurring to sensitive life stages of fish such as juvenile and larval fishes or to aspects of populations or communities not yet being assessed. Effects may also be occurring to trophically linked organisms, such as the bald eagle or river otter, due to the biotransfer ability of some constituents. The SERA provided the understanding and justification for formulating the assessment endpoints associated with the next phase of ecological evaluation for the IOUs.

557 **Transport of Select Man-made Radionuclides in the Upper Sun-basin of the Savannah River.** Fjeld, R. A., Clemson University; Reboul, S. H., Westinghouse Savannah River Company; Elzerman, A. W., Clemson University. Field measurements were conducted to determine temporal and spatial distributions of select man-made radionuclides, principally ^{3}H and ^{137}Cs, in the upper sub-basin of the Savannah River including Lakes Keowee, Hartwell, Russell, and Thurmond. Included in the study are measurements of radionuclide concentrations in the dissolved phase, in suspended particulate matter, and in bottom sediment. The data are used to develop a conceptual model for the transport and fate of radionuclides released to the system from the Oconee Nuclear Station. A compartmental mathematical model for transport in Lake Hartwell is developed, and model predictions are compared to field data.

558 **The Use of Lead-210 Dating to Determine Sedimentation Rates in the Twelve Mile Creek Region of Lake Hartwell.** Coates, J.T.*, DeVol, T.A., EE&S, Clemson University, Richardson, P.A., CE, Clemson University, Clemson, SC. 29634-0919 In order to analyze and manage an ecosystem it is often necessary to determine the rates at which certain processes change. Lake Hartwell is one ecosystem that has been studied with some degree of frequency over the past several years. In the 1970's it was found that a capacitor manufacturer contaminated aquatic life and sediments in Lake Hartwell with polychlorinated biphenyls (PCBs) as a result of discharges into Twelve-Mile Creek. Numerous sediment samples collected since the early eighties indicate significant levels of PCB contamination, on the order of 10^{2}-20^{2} μg/gm and sediment deposition rates between 0.5 and 6.0 cm per year. Deposition rates have historically been measured by monitoring the PCB concentration maxim versus depth. Sediment resuspension events in the late eighties have raised question as to the validity of this technique to determine deposition rates. Lead-210 based sediment dating was first introduced by E.D. Goldberg in 1963 and has gained popularity over the over the past thirty-five years and was used in this investigation to obtain record of sediment age in order to substantiate previous measurements. Lead-210 dating using X-ray spectrometry correlates well with sediment deposition rates calculated from PCB measurements in the soil column in areas where resuspension events were minimal. Two radionuclide measurement techniques, direct X-ray assay using a Si(Li) X-ray spectrometer and Cerenkof counting of the high energy beta emitting progeny of lead-210, bismuth-210 were compared during this investigation.

559 **Lipid Composition of Suspended Particulate Organic Matter in Coastal Plain Blackwater Tributaries of the Savannah River.** Mills, G.L.*, Wolfe, C. and McArthur, J, Savannah River Ecology Laboratory, Aiken, SC. Suspended particulate organic matter (SPOM) was collected seasonally from 3[rd] and 5[th] order sites in a blackwater stream tributary of the Savannah River in Aiken County, South Carolina. Fatty acids, alcohols, and hydrocarbons (HC) were determined in the isolated SPOM. Fatty acids were the most abundant component among the lipid classes examined with concentrations ranging from 8.5 - 60.2 $\mu g\,L^{-1}$. Carbon chain-lengths ranged from C_{12}-C_{32}. The qualitative assemblage of major fatty acids identified was the same at both sites. The 16:0, 16:1•9, 18:0, 18:1•9, and 18:2•9,12 fatty acids predominated while significant concentrations of the even-chained saturated components C_{24}-C_{30} derived from cuticular plant waxes were also found. Microbially derived iso and anteiso methyl branched-chained and ß hydroxy fatty acids were also present. Aliphatic alcohols concentrations ranged from 0.52-2.73 $\mu g\,L^{-1}$ and was comprised of normal even-chained components with carbon chain-lengths between C_{16}-C_{30}. The qualitative distribution of these aliphatic alcohols was the same at both sites and was dominated by the higher molecular weight compounds (C_{22}-C_{30}) derived primarily from cuticular plant waxes. In general, lipid concentrations at the 3[rd] order site generally had higher and more variable concentrations of the lipid classes compared with the 5[th] order site. The ratio of saturated to unsaturated fatty acids was higher at the 3[rd] order site indicating that the SPOM at this site was of more recent origin and less decomposed. The ratios of cuticular to noncuticular fatty acids and alcohols support this conclusion. These results indicate an export of particulate lipids of higher carbon resource quality from upstream to lower stream reaches.

560 **Long-term Ecological Decay Rates for Cesium-137 in Fish from Contaminated Streams and Reservoirs in the Southeastern USA.** Paller, M.H.*, and Littrell, J.W., Savannah River Site, Aiken, SC. We computed ecological decay rates for cesium-137 in several types of fish from two reservoirs and three streams on the Savannah River Site (SRS), a former nuclear materials production facility. Our computations reflected long-term (approximately 1970-1996) reductions in

cesium-137 levels in whole fish following radionuclide releases that occurred primarily in the 1960s. Ecological half-lives for fish from two of the streams (3.2-4.9 yrs) were significantly shorter than for fish from the reservoirs (7.5-10.1 yrs), probably because of the continuous export of cesium-137 from the streams. A half-life calculated for fish from a third stream, which received nearly all of its discharge from one of the reservoirs, was comparable to the reservoirs (10.8 yrs). All decay rates were more rapid than expected on the basis of physical isotope decay alone (half life of 30 years). Ecological decay rates in fish were strongly correlated with ecological decay rates in water. Individual taxa from the same locations differed in mean levels of cesium-137 but not in cesium-137 decay rates. We estimated when cesium-137 concentrations in fish would decline to levels safe for human consumption based on conservative risk assumptions. Our results indicated that contamination was more persistent in the reservoirs and that the time needed to reach safe levels was more strongly related to the ecological decay rate than to the initial cesium-137 concentration.

561 Use of the Sediment Quality Triad, TIEs, and Gradient Studies in Identifying Toxic Hotspots in San Francisco Bay. Hunt, J. W.*, Anderson, B. S., Phillips, B. M., Tjeerdema, R. S., University of California, Santa Cruz, CA; Fairey, R., Moss Landing Marine Laboratories, Moss Landing, CA; Puckett, H. M., Stephenson, M., California Department of Fish and Game, Granite Canyon, CA; Taberski, K. M., San Francisco Bay Regional Water Quality Control Board, Oakland, CA. After screening 127 sites in San Francisco Bay for sediment toxicity, the California Bay Protection and Toxic Cleanup Program revisited 25 sites for evaluation with the sediment quality triad, incorporating sediment chemical, toxicological, and benthic ecological measures. At many sites, samples were collected along suspected contamination gradients, and sediment TIEs were conducted at three sites. Sediment toxicity was assessed using amphipods in homogenized sediment and sea urchin embryos exposed to intact sediment cores at the sediment-water interface. Toxicity tests responded to increasing contamination along gradients, as indicated by sediment chemistry guideline (ERM) summary quotients and by concentrations of individual chemicals. A Relative Benthic Index corresponded with chemistry and toxicity at many sites, as did individual benthic community metrics. TIEs implicating trace metals corroborated bulk-phase chemical measurements above guideline values, and corresponded with patterns of response by different toxicity test species in solid-phase tests. High concentrations of anthropogenic contaminants and hydrogen sulfide were related to biological impacts at many sites.

562 Using Field Survey Data to Predict Exposure Scenarios and Interpret Toxicity Test Results. Watzin, M.C., University of Vermont, School of Natural Resources, Burlington, VT. The search for ways to make toxicity test data more meaningful has led to a variety of approaches to integrated sediment assessment. Multivariate methods are one way to find associations between measured characteristics of the benthic environment and the macroinvertebrate community. Using multiple regression modeling on a series of data collected from Lake Champlain, Vermont, we found that sediment grain size distribution was as important or more important than contaminant concentration in explaining benthic community composition and toxicity test results. Although not demonstrating causal linkages, these statistical analyses suggested which factors might be important in driving organism responses. Field surveys can also provide information about the *in situ* benthic environment. When environmental characteristics known to affect bioavailability (such as SEM/AVS ratio and percent organic matter content) are known, control sediments or other media can be modified to provide comparisons that more realistically represent field exposure scenarios. In TIEs using a pore water test, we added DOC (sodium humate) to reconstituted water to more realistically mimic field media in confirmatory tests. Whole sediment controls can also be selected or adjusted using formulated sediment constituents. By tailoring techniques in these ways, laboratory toxicity testing might provide better prediction of field responses.

563 Determining the Effect of Ammonia at Complex Sites: Laboratory and *In situ* Approaches. Burton, G. A., Jr., Rowland, C., Kroeger, K., Greenberg, M., Lavoie, D., and Brooker, J., Institute for Environmental Quality, Wright State University, Dayton, OH. An assessment of ammonia toxicity was conducted using *Hyalella azteca, Pimephales promelas, Chironomus tentans, Corbicula fluminea, Hydra attenuata,* and *Lophopodella carteri* in laboratory and *in situ* exposures. Results were compared with indigenous benthic macroinvertebrate and fish communities. Warm waters (> 30 C) were found to increase ammonia concentrations and toxicity. In addition, warmer waters accentuated the toxicity of water and sediment related toxicity. Toxicity due to ammonia varied between species and test media. Laboratory exposures showed decreasing ammonia in site sediment exposures through time, however other treatments showed increasing ammonia. The approach allowed for evaluations of the role of different sources of ammonia, thermal interactions, and showed differences between laboratory and field results. Both laboratory and field techniques were necessary to determine the significance of the various stressor effects.

564 Ammonia Toxicity to Amphipods in 4-d and 10-d, Water Only and Spiked Sediment Tests. Kohn, N. P.*, Battelle Marine Sciences Laboratory, Sequim, Washington; Swan, B. K., Michigan Technical University, Houghton, Michigan; Gruendell, B. D., Battelle Marine Sciences Laboratory, Sequim, Washington. Bioassays were conducted to answer the questions: Does the presence of sediment moderate ammonia toxicity to amphipods? Is the 4-d water only dose-response a valid and accurate predictor of 10-d sediment toxicity? 4-d water only and 10-d spiked sediment bioassays were conducted with the amphipods *Ampelisca abdita, Rhepoxynius abronius,* and *Leptocheirus plumulosus*. These three amphipod species are commonly used to evaluate marine and estuarine sediment toxicity. 4-d spiked sediment and 10-d water only tests were also conducted with *R. abronius* and *L. plumulosus* to evaluate the effect of presence or absence of sediment on ammonia dose-response. Each test had four or five test concentrations and a control. Native control sediment for each species was used for the spiked sediment tests. In water only and spiked sediment tests of the same duration, ammonia toxicity to *R. abronius* and *L. plumulosus* was moderated by the presence of sediment. The dose-responses for 4-d water only and 10-d spiked sediment tests were comparable for all three species: NOECs were similar in both exposure scenarios, but the dose-response was steeper in the 10-d spiked sediment test than in the 4-d water only test, resulting in lower spiked sediment LC50s. The close relationship between 4-d water only and 10-d spiked sediment dose-response, especially at lower concentrations, validates the use of NOECs derived from 4-d water only tests. At higher concentrations, the water only dose-response might under predict ammonia toxicity in a 10-d sediment test.

565 Sediment versus Overlying Water as Sources of Metals to Benthic Tube-dwellers: Evidence from Oxygen Profiles in Their Microenvironments. Wang, F., Tessier, A., and Hare, L., INRS-Eau, Université du Québec, Québec, Canada. Current strategies for metal-contaminated sediment management presume that benthic animals take up most of their metals from contaminated sediments. Although this has been generally observed in laboratory experiments, field studies in two Canadian lakes demonstrated that most benthic tube-dwellers took up their cadmium almost exclusively from the overlying water. To explain this phenomenon, we hypothesized that conditions in the microenvironment that animals create resemble more those in overlying water than those in the surrounding sediment. To test this hypothesis, we measured O_2 concentrations in and around the tubes of the insects, *Sialis velata* (Megaloptera) and *Hexagenia limbata* (Ephemeroptera), with oxygen microelectrodes. Oxygen profiles in the sediments and across the tubes were measured with a spatial resolution of 25 μm, whereas fluctuations in O_2 concentrations in the tubes were measured with a temporal resolution of 10 sec. at temperatures of 4, 10 and 20 °C. Oxygen profiles reveal high O_2 concentrations (50-100 % of air saturation) in the animals' tubes in otherwise anoxic sediments. Temporal patterns in oxygen concentrations inside the tubes suggest that the mayfly *H. limbata* irrigates its burrow continuously, whereas irrigation by the alderfly *S. velata* is periodic and varies in rate with temperature. Because both animals irrigate their tubes with oxygenated overlying water and hence live in an oxic microenvironment of their own making, our results support that overlying water could be an important source of metals to these tube-dwellers. Furthermore, the thin layer (1-2 mm) of oxic sediment created by the animals around their tubes is likely to influence sediment and porewater metal chemistry.

566 Importance of Burrowing Behavior in Predicting Cadmium Accumulation and Toxicity to Benthic Animals. Hare, L.*, Tessier, A., INRS-Eau, Sainte-Foy, Québec, Canada. Given the importance of designing reliable, theoretically sound, models to assess the impact of sedimentary trace metals, we carried out an *in situ* experiment to determine the major environmental compartment, sediment or overlying water, from which benthic animals take up Cd in nature. We used a spiked sediment bioassay approach that involved creating a Cd concentration gradient in the sediment compartment without altering Cd concentrations in the overlying water compartment. Sediment was collected from the littoral zone of two lakes, Cd was added, and the sediment replaced in trays on the lake bottoms to be colonized by benthic animals. Cadmium concentrations in a few colonizing taxa, including the crustacean *Hyalella azteca*, were not related to the sediment and interstitial water Cd gradient suggesting that they take up their metal from the overlying water compartment. At the other extreme, tubificid oligochaetes, the only taxon on which toxic effects were measured, appeared to take up almost all of their Cd from the sediment compartment, likely a result of their head-down, non-irrigating, feeding behavior. Cadmium concentrations in the remaining taxa studied (including *Chironomus* and *Hexagenia*) represented a mixed response, i.e., Cd appeared to be accumulated from both the overlying water and the sediment compartments. The importance of the overlying water compartment as a Cd source for many of these benthic animals is likely a consequence of their behavior. From X-ray images we determined that most burrowing insects live in U-shaped tubes, and measurements with oxygen micro-electrodes indicated that animals maintain an oxic micro-environment in their tubes. Our experimental results suggest that the behavior of insects explains the absence of a toxic effect on these animals living in our highly Cd-contaminated sediments.

567 Effect of Exposure Source (Sediment Vs. Food) on the Uptake and Toxicity of ^{14}C-DDT to the Marine Polychaete *Neanthes arenaceodentata*. Lotufo, G.R.*, Farrar, J.D., AscI Corporation, Vicksburg, MS; Bridges, T.S., Moore, D.W. USACE Waterways Experiment Station, Vicksburg, MS. The relative importance of sediment and diet as contaminant sources of uptake and toxic effects was examined. *Neanthes arenaceodentata* was exposed to a concentration series of either a DDT- spiked sediment (SS), a DDT-spiked food (SF), or to both media simultaneously (SS/SF). Measured DDT sediment concentrations were 41, 90, 177, and 374 μg/g dry wt. Worms were fed Tetramarin. DDT concentrations in the food equaled the sediment concentrations on an organic carbon basis. Emerging juveniles were exposed to DDT for 28 days. Survival was not significantly reduced in any exposure scenario. DDT, however, caused a significant decrease in growth. Detrimental effects were much stronger with a dietary exposure to DDT. Worms were significantly smaller at the highest concentration (374 μg/g) in SS. In the SF and SS/SF exposure scenarios, however, growth was significantly reduced at 90 μg/g. Tissue concentrations of DDT and its metabolites at day 28 were determined. Efficient biotransformation of DDT to DDE might have contributed to the high tolerance to DDT. At any given treatment, DDT tissue concentration was highest in worms exposed to both spiked sediment and food and lowest in worms exposed to spiked sediment only. The exposure source of contaminants had a strong effect on both the uptake and toxicity of DDT. Dietary uptake yielded higher body residue and stronger adverse effects compared to sediment uptake.

568 Inter-laboratory Evaluation and Comparative Sensitivity of a Chronic Sublethal Sediment Bioassay Using *Neanthes arenaceodentata*. Bridges, T.S.*, USAE Waterways Experiment Station, Vicksburg, MS; Farrar, J.D., AScI Corp., Vicksburg, MS; Gardiner, W.W., Niewolny, L.A., Battelle Marine Sciences Laboratory, Sequim, WA. The precision and sensitivity of bioassays must be evaluated prior to regulatory implementation. Variability in test results for a 28-day sediment bioassay using the polychaete *Neanthes arenaceodentata* was evaluated during an inter-laboratory evaluation involving six laboratories. The bioassay is initiated with <7 day old juveniles. Worms are fed ground TetraMarin (2 mg/worm) and alfalfa (1 mg/worm) twice a week during the 28-day exposure. The test endpoints are survival and growth. During the inter-laboratory evaluation worms were exposed to a dilution series (0, 6, 12, 25, 50, 100 %) of contaminated sediment collected from Black Rock Harbor (BRH). Survival effects were only observed in the 50% and 100% BRH treatments. Strong and consistent exposure-response relationships were observed for the growth endpoint. The LOECs for the growth endpoint were 6% BRH (3 labs), 12% BRH (2 labs), and 25% BRH (1 lab). The sensitivity of this bioassay was evaluated using the BRH dilution series in side-by-side comparisons with a 56-day version of the same protocol, a 28-day bioassay using the amphipod *Leptocheirus plumulosus*, and the 20-day *Neanthes* protocol used in the Puget Sound Estuary Program (PSEP). Results were similar for the 28- and 56-day *Neanthes* bioassays and the amphipod chronic test. Results were quite different for the 20-day PSEP protocol where no treatment effects were observed on survival or growth. The results of this study demonstrate that the 28-day *Neanthes* bioassay will provide both sensitive and precise measures of sediment toxicity.

569 Relative Sensitivity of Endpoints Measured in Long-term Water-only Exposures with the Amphipod *Hyalella azteca* and the Midge *Chironomus tentans*. Kemble, N.E.*, Dwyer, F.J., Ingersoll, C.G., Schuerenberg, H. D. Environmental and Contaminants Research Center, U.S. Geological Survey, Columbia, MO. Endpoint sensitivity ss being evaluated in 42- to 60-day water-only exposures with *Hyalella azteca* and *Chironomus tentans* to cadmium, ammonia, and 4-nonylphenol. Endpoints evaluated included survival, growth (length and weight), and reproduction (number of young/female). Endpoints in the midge exposure included survival, growth, emergence, and egg hatching. These studies will determine if there is a substantial increase in endpoint sensitivity in long-term tests compared to the 10-day sediment tests where lethality and growth are measured. A control and 5 exposure concentrations (50% dilution series) were used in each study. Concentrations for each chemical were determined using a 10-day range finder study. For the amphipod tests a total of 24 replicates were used in each test, (4 replicates for 10-day growth and survival, 4 replicates for 28-day growth and survival and 16 replicates for 28- to 42-day reproduction, growth and survival (8 replicates for continual chemical exposure and 8 replicates into clean water on Day 28)). Continual chemical exposures and clean water exposures will provide guidance for selecting procedures to conduct sediment reproduction studies with *H. azteca*. For the midge test, a total of 16 replicate were used in each test, (4 replicates for 20-day survival and growth, 8 for emergence, and 4 for production of auxiliary males (set up on Day 10)). Results to date indicate measurement of sublethal growth in 10- to 60-day exposures with amphipods and midges provides unique sublethal information. In addition, reproduction was generally reduced only at exposure concentrations that also reduced growth.

570 A Comparison of Test Endpoints in Sediment Toxicity Tests with *Chironomus tentans* and *Hyalella azteca*. Leppanen, C.J.*, University of Maine, Orono, ME; Maier, K.J., University of Memphis, Memphis, TN. Test endpoints survival, weight, and length were compared for sensitivity and consistency in sediment toxicity tests performed with *Chironomus tentans* and *Hyalella azteca*. Exposure sediments were collected from urban sections of the Wolf River adjacent to, upstream from, and downstream from a CERCLA site; reference sediments were collected from the rural headwaters of the Wolf River; control sediments were formulated. Whole sediment toxicity tests were conducted for 10 days with reconstituted water renewal. Sediments were screened for 17 organochlorine pesticides and PCBs; none of these contaminants were detected in sediments. Al, Ba, Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn were detected in some significant concentrations in urban river sediments. Sediments collected from the three urban reaches of the Wolf River demonstrated toxicity with reduced survival and growth in both *C. tentans* and *H. azteca* when compared to reference sites and controls. Utilizing the three comparative endpoints showed highly consistent results within and among species, a strong correlation between the weight and length of surviving invertebrates, and an increased sensitivity in results using length as an endpoint. In situations where survival used as an endpoint is not sensitive, the growth endpoints (length and weight) might be more helpful in quantifying toxicity.

571 **The Ecological Risk Assessment Process: Have We Lost Our Perspective?** Meyers-Schöne, L.*, IT Corporation, Albuquerque, NM. This paper provides the introduction and background to the session on "Strengthening the Ecological Risk Assessment Process". Recent criticisms of current methodologies in ecological risk assessment have focused on specific aspects of general screening level assessment procedures at the exclusion of all other areas of ecological risk assessment. Commonly, these criticisms relate to extrapolations of literature-obtained toxicity data, utilization of uncertainty factors, body weight scaling, and estimation of ingestion rates. Current debates related to these topics indicate a loss of perspective with too much emphasis placed on the screening assessment, which is only a tool to conservatively estimate ecological risk in the absence of site-specific data. The intent of this paper is to provide a broader perspective for recent criticisms of the ecological risk assessment process. There is general agreement that weak points exist within the screening process. However, its relevance to the bottom line or overall goal of an ecological risk assessment as performed under the Comprehensive Environmental Response, Compensation and Liability Act, Resource Conservation and Recovery Act, and National Environmental Policy Act should be emphasized. Concerns of the ecological risk assessment process need to be refocused toward areas of greater significance- the assessment of ecological risks at the appropriate scale using assessment and measurement endpoints that are relevant to ecological concerns. Areas of needed research and development within this area, include ecological prioritization, ecological resource values, and watershed/landscape level concerns, which are the focus of papers within this session.

572 **Risk Assessment and Risk Management: EPA's Perspective.** Sergeant, A.*, Barton, A., U.S. EPA, Washington, DC; Wharton, S. U.S. EPA, Kansas City, KS. Ecological risk assessment is a rapidly evolving field, and both risk assessors and risk managers are finding themselves in new roles. We will present EPA's recent efforts to clarify those roles, particularly in the area of identifying which ecological values and entities should be protected. The presentation will address how to identify ecological risk-management goals and objectives, what to protect, selection criteria and a process for applying them in specific EPA Programs, and a process for identifying additional items for protection. It will specifically focus on a document prepared for the "bookshelf" of resources to support implementation of EPA's *Guidelines for Ecological Risk Assessment*. This document will be a tool for risk assessors and risk managers to use during the planning process--when reviewing the information available before a risk assessment is initiated, identifying the risk-management decision that will eventually be made, and deciding what questions need to be investigated to support that decision.

573 ***Quo vadis* Ecological Risk Assessment.** Suter, G. W., Oak Ridge National Laboratory, Oak Ridge, TN. Ecological risk assessment has been practiced for over a decade. It now employs a large fraction of SETAC members and has achieved sufficient prominence to be regularly attacked by critics. However, it has not achieved the influence or respectability as a practice that human health, engineering or financial risk assessment have achieved. In part this is because there are no accepted practices. This state can no longer be blamed on the relative youth of our field, or even the complexity of ecosystems. However, those considerations make the usual standardization processes too cumbersome. One approach to solving this problem is being developed for the U.S. Department of Defense. It consists of a framework for ecological risk assessment of training and testing programs that is designed to incorporate specific assessment tools. The needed tools will be provided by the assessors who use the framework. In that way the military assessment community will teach itself how to best assess risks from tank traffic, underwater explosions, aircraft noise, etc. The development process would be equivalent to learning a language with gradual progression from a few poorly pronounced words and phrases to fluent conversation or the evolutionary development of the eye from light-sensing cells to image-forming organs. Such developmental systems have been termed schemata. A schema for ecological risk assessment in general would allow us to share what we know, evaluate the quality of the knowledge, identify what we don't know, and advance the practice. This work was supported by a grant from the Strategic Environmental Research and Development Program.

574 **Linking Ecological Understanding and Ecological Risk Assessment: The DQO Process.** Pye, L.H., Woodward-Clyde Consultants, Denver, Colorado; Current ecological risk assessment procedures guidance is principally designed to support quantitative risk characterization of individual species as receptors and surrogates for population, community, and ecosystem-level effects. Developing a work plan for investigating ecosystem-level changes resulting from large-scale contaminant release requires a decision process to address risk at various ecological organizational levels and to address risks posed by the potential remedy options. Employment of an landscape approach provides a practical means to define the structural and functional components of a heterogeneous environment. Landscape ecology also provides a sensible, analytic approach and practical basis for identifying the principal study questions and broad spatial scale, risk analysis decisions that require data to address the contamination problem. Some of the most costly, wasteful efforts associated with large-scale, landscape-level ecological risk assessments arise when the remedial investigation data collection programs are not specifically linked early-on to the risk managers' and stakeholders' ecological information needs. Such needs include identifying nature and extent of the ecological threat (versus nature and extent of contamination) and identifying potential remedy risks posed by habitat conversion, habitat fragmentation, and exotic species introduction. The data quality objectives process is a useful means to link these large-scale ecological considerations with ecosystem management and remedy selection concerns during the remedial investigation and in the initial planning, problem formulation stage of the ecological risk assessment.

575 **Three Types of Background and Their Uses in Ecological Risk Assessment.** Burns, T.P.*, Cornaby, B.W., Hadden, C.T., Mitz, S.V., and Ryan, P.F., Science Applications International Corporation, Oak Ridge, TN. We define three types of background and three uses for background chemical data in ecological risk assessments (ERAs) at hazardous waste sites (HWSs). Assume all background locations have geology and ecology similar to the HWS. Local background is nearby, upgradient of the HWS with respect to major transport pathways, possibly downgradient by minor pathways. Local background and the HWS potentially share site-specific, regional, and global sources of contaminants. Regional background is not impacted by the HWS, but is potentially impacted by human activities (sources) common to both locations; they share regional and global sources. Natural background is not impacted by either the HWS or regional sources, but may have globally distributed contaminants. All three background types can be used in ERAs to identify site-related contaminants (SRCs), identify contaminants of concern (COCs), and develop remedial goal options (RGOs). Distinguishing site-specific sources from other sources is important for identifying SRCs. Identifying COCs from among regionally or globally distributed contaminants on the basis of incremental vs. total risk assumes that receptors are not at unacceptable risk at

background. Local and regional background levels have implications for what are safe and practical RGOs. The pros and cons of each background type for each use are evaluated from the perspective of three end users: responsible party, risk assessor, regulator. We compare possible rankings of background types for each use for each of the three end users. Rankings for identifying SRCs and developing RGOs are potentially opposite for responsible parties and regulators because of conflicting goals, e.g., minimize cost vs. maximize protection. Regional background may be the compromise choice for SRCs and RGOs and the first choice for identifying COCs.

576 Wildlife Toxicity Data and Ecological Risk Assessment: Problems and Solutions. Sample, B.E.*,Rose, K.A., Suter, G.W., II, Arenal, C.A., Oak Ridge National Laboratory, Oak Ridge, TN. The current approach for risk assessment for wildlife consists of comparison of contaminant exposure estimates for individual animals to literature-derived toxicity test endpoints. These test endpoints are assumed to estimate thresholds for population-level effects. For several reasons, uncertainties associated with this approach are considerable. First, because toxicity data are not available for most potential wildlife endpoint species, extrapolation of toxicity data from test species to the species of interest is required. There is no consensus on the most appropriate extrapolation method. Second, toxicity data are represented as statistical measures (e.g., NOAELs or LOAELs) that provide no information on the nature or magnitude of effects. The level of effect is an artifact of the replication and dosing regime employed, and does not indicate how effects might increase with increasing exposure. Consequently, slight exceedance of a LOAEL is not distinguished from greatly exceeding it. Third, the relationship of toxic effects on individuals to effects on populations is poorly estimated by existing methods. It is assumed that if the exposure of individuals exceeds levels associated with impaired reproduction, then population level effects are likely. Uncertainty associated with this assumption is large because depending on the reproductive strategy of a given species, comparable levels of reproductive impairment may result in dramatically different population-level responses. We are working on several tasks to address these problems: 1) investigation of the validity of the current allometric scaling approach for interspecies extrapolation; 2) development of dose-response models for toxicity data presented in the literature; and 3) development of matrix-based population models that, coupled with dose-response models, will allow for realistic estimation of population-level effects for individual responses. Results of the first year's efforts will be presented.

577 Population Level Risk Assessment: An Innovative Approach to Assessing Ecological Risk. Stirling, B.A.*, Oregon DEQ, Portland, OR; Hope, B.K., Oregon DEQ, Portland, OR; Chew, C.M., Roy F. Weston, Inc., Seattle, WA; Peterson, J.A., EMCON, Eugene, OR. In 1995, Oregon enacted amendments to its state hazardous waste site cleanup law which emphasize risk-based remedial action decisions. In a departure from U.S. EPA practice, the amended statute and associated rules require that protection of ecological receptors occur at the population-level for all plants and animals not listed as threatened and endangered. By rule, the acceptable risk level for populations of ecological receptors is a 10 percent chance, or less, that 20 percent or more of the total local population would receive an exposure greater than the toxicity reference value for a hazardous substance. We describe a practical approach for performing population-level ecological risk assessments using a combination of relatively simple techniques and present a case study in which this approach has been applied. The approach involves: (1) establishing a distribution of exposures and a contaminant-specific toxicity reference value, either as a point value or a distribution, for an individual receptor, (2) estimating population abundance of these receptors within their site- and species-specific local population boundary, (3) estimating the probability of an individual receptor experiencing an exposure in excess of the toxicity reference value, (4) estimating the number of individual receptors in the local population likely to experience an exposure above the toxicity reference value greater than 10% of the time, and (5) determining whether this number is greater than 20% of the total local population.

578 Screening Ecological Risks at Superfund Sites by Comparing Exposed Receptors' Tissue Residues to Background Burdens from Reference Areas. Henningsen, G.*, U.S. EPA, Denver, CO; Wickstrom, M., Landcare Institute, Christchurch, New Zealand; Rimar, B., U.S. EPA, Denver, CO; Everett, S., UDEQ, Salt Lake City, UT; Dorward-King, E. J., Kennecott Utah Copper, Magna, UT; Graham, R., U.S. EPA, Denver, CO; Hoff, D. J., U.S. EPA, Denver, CO. Ecological risk assessments at Superfund sites and other areas pose challenges for determining exposures to stressors as well as the risks of effects (e.g., toxicologic) from those stressors when receptors are over-exposed. An effective scientific approach that serves risk assessors well in a tiered or screening framework involves the use of stating a null hypothesis regarding exposure to potential stressors. EPA Region VIII has successfully used this approach to biostatistically screen-out or screen-in the potential for excess ecological risks related to Superfund sites. EPA regional BTAGs (biological technical assistance groups) have been employed to design representative sampling and analyses, based upon exposure units, of appropriate tissues from site-exposed receptors and from background-exposed receptors at comparable reference areas and times. The premise is (with adequate study design) that if there are no significant elevations of stressors in exposed vs reference receptors, then there can not possibly be any excess risks associated with site sources. In such cases, risk investigations can stop and the risk assessment finished for that scenario. If over-exposure to stressors occurs in respect to references, then studies can proceed to subsequent tiers for exploring nature/extent, magnitude of responses, or mechanisms; and/or possibly to further screening of stressor effects. EPA and others have been developing risk-based screening benchmarks for this use, such as with the examples of sediment and soil-screening toxicant concentrations.

579 Benefits of Watershed and Landscape Approaches to Ecological Risk Assessment to Support the Remedial Decision Process. Burris, J., ISSI, Inc., Denver, CO. Recent trends in ecological risk assessment place an emphasis on conservative screening level methods. This emphasis is at the expense of appropriate scale assessments using endpoints relevant to ecological concerns and management goals. The benefits of using watershed/landscape approaches in ecological risk assessment for the support of remedial decision making will be explored in the presentation of two case studies. One case study concerns heavy metals in soils and the second heavy metals and chlorinated organics in sediments. For each case study, the presentation will examine the selection of appropriate site-specific endpoints, the assessment approach and methods, results and decision making. The watershed/landscape approach will be evaluated in comparison to the "screening" approach. Questions to be examined include: Should conservative screening exercises become the basis of definitive decision-making assessments? Or define the scope of population or community level studies? Should assessments focus/ rely on field measurements? What are the priorities and goals for ecological protection in each type of assessment? What adverse effects are considered "significant"? Does natural resource damage assessment influence the scale of assessments and the definition of significant? And how does each assessment methodology consider the potential remedial options. Potential areas of research and development are identified.

580 Integration of Habitat Considerations and Residual Risk in Selecting Remedial Alternatives. Durda, J.L.* and Preziosi, D.V., THE WEINBERG GROUP INC., Washington, DC. During the past few years, there has been a growing recognition that clean-up of chemical contamination can, in certain situations, do more environmental harm than good. For example, dredging operations designed to remove contaminated sediments can destroy benthic habitats, resuspend and redistribute contaminated sediments, disrupt surface hydrology, and change sedimentation patterns. We were confronted with a situation in a southern cypress-hardwood swamp, where removal of pesticide contamination in wetland sediments would have necessitated clear cutting and dredging of extensive tracts of mature forest and patches of early successional herb and shrub habitat in previously cleared areas. Leaving the pesticide contamination in place would have preserved habitats, but not changed the baseline risk levels, which were regarded as unacceptable by regulatory agencies. To evaluate remedial alternatives, we conducted a series of analyses that were designed to assess the net ecological benefit of clear cutting and dredging in different portions of the flood plain. Our analysis considered four factors: (1) ecosystem functional values (e.g., as foraging or breeding area); (2) habitat uniqueness and prevalence; (3) habitat restoration potential; and (4) residual risk. Based on these analyses, the recommended remedial alternative consisted of dredging of sediments in early successional herb and shrub habitat, and in portions of the cypress swamp containing the highest pesticide concentrations. Risk-reduction was disproportionately larger than the size of the habitat remediated and less than 10% of the total habitat of the flood plain was disturbed. In addition, the approach helped ensure that remediation would improve overall ecological conditions at the site and that dollars and time were not unnecessarily spent.

581 Green Chemistry - Designing Chemical Products and Processes for the Environment. Green Chemistry, or chemistry designed to reduce or eliminate the use or generation of hazardous materials associated with the manufacture and use of chemical products, has achieved remarkable successes in academia and industry. Through the use of Green Chemistry, chemists have developed ways of removing millions of pounds of hazardous substances from the products and processes that society needs, without sacrificing scientific innovation and creativity. This presentation will provide an overview of the various programs that are promoting green chemistry through education, research, and recognition. This presentation will provide an overview of the various technical areas of Green Chemistry that are being researched, developed, and implemented in academia and industry and will highlight their resulting human health and environmental benefits.

582 Genesis of Durable Hydrodechlorination Catalyst for Selective Conversion of CCl_4 to $CHCl_3$ and Potential Commercial Application. Conrad Zhang, Bruce Beard. Due to its high potency in depleting the stratosphere ozone layer, the production and use of carbon tetrachloride (CCl_4) has been phased out together with chlorofluorocarbons in developed countries according to the Montreal Protocol effective as of January 1 1996. Being itself much less destructive than CCl_4 for stratosphere ozone depletion, chloroform ($CHCl_3$) is used in the manufacture of replacement HCFC's. However, in the production of $CHCl_3$, CCl_4 is still produced as a byproduct. The disposal of this harmful byproduct, typically by incineration, has become an environmental challenge and a major economic burden to the manufacturers of $CHCl_3$. Hydrodechlorination of CCl_4 to $CHCl_3$ is an economically and environmentally attractive alternative for CCl_4 disposal. Effort dating back to the early seventies as well as recently have succumbed to catalyst deactivation or heavier chlorinated by-product formation under practical reaction conditions. A novel catalyst pretreatment will be reported that renders commercially available Pt/Al_2O_3 pellet catalysts durable under commercially viable operating conditions. While the untreated catalyst were rapidly deactivated during reaction (<1 hour), the treated catalyst has been demonstrated to give excellent performance for over 2,000 hours without deactivation. A commercially viable process has been developed based on this catalyst technology.

583 Quantitative Structure Activity Relationships as Pollution Prevention Tools in the Product Development Cycle. Ewell, W.S. and Ruffing, C.J., Eastman Kodak Company, Rochester NY, USA. The design of safer chemicals requires that health, safety and environmental (HSE) properties be considered early in the product development process. The consideration of HSE attributes in early chemical design is often done in the absence of test data. Professional judgment and quantitative structure activity relationships (QSAR) are commonly used in early assessments. The US EPA has made available a Pollution Prevention Framework, that allows the estimation of many environmental fate and effect parameters. The use of QSARs in Kodak early assessment systems is described. Several examples showing how early assessment led to focused research, cost savings and pollution prevention will be discussed.

584 Estimation Tools for Designing Safer Chemicals - Determining Persistent, Bioaccumulative, and Toxic (PBT) Chemicals. Howard, P.H.*, Syracuse Research Corporation, N. Syracuse, NY; Boethling, R.S., U.S. EPA, Washington, DC. When reviewing new chemicals or selecting an alternative chemical, investigators need to consider the PBTness of a chemical. Often experimental data are not available and one needs to rely on estimates. A review of computer programs and other techniques that are available for selecting safer chemicals will be presented with focus on fate and ecotoxicity. Latest criteria for PBT, Persistent Organic Pollutants (POPs), and Persistent and Long Range Transport (PLRT) chemicals will be considered.

585 Designing Biodegradable Chemicals. Boethling, R.S., U.S. EPA, Office of Pollution Prevention and Toxics, Washington, DC. Under the Pollution Prevention Act it is the policy of the United States that pollution should be prevented or reduced at the source whenever possible. One way to accomplish this is to design safer chemicals. Chemicals that persist in the environment remain available to exert toxic effects and may bioaccumulate. Since microbial degradation is the major loss mechanism for most organic chemicals in soil, water and sewage treatment, biodegradability should be included as a factor in product design along with product efficacy, economics, etc. Biodegradability has been an important design consideration for down-the-drain products like laundry detergents for decades, but not for chemicals with mainly non-consumer uses. Yet the relationship between molecular structure and biodegradability is now understood well enough to be broadly applicable. It is *essential* to extend the "benign by design" concept to commercial chemicals generally, because i) we cannot know in advance all possible toxic effects of released chemicals; ii) production and release of a substance may increase significantly, with unforeseen consequences, if a chemical is successful in the marketplace; iii) new uses may develop over time, also affecting exposure; iv) even if local controls are thought to be adequate, chemicals may be exported to other nations with less stringent environmental controls. It is shown by means of examples, drawn from high-volume chemicals in current use, how enhanced biodegradability has or *might* have avoided unnecessary environmental damage.

586 Prediction of Biodegradability in the MITI-I Test Using Fragments from the BIODEG Program. Howard, P.H.*, Syracuse Research Corporation, N. Syracuse, NY; Boethling, R.S., U.S. EPA, Washington, DC.; Loonen, H., RIVM, Bilthoven, The Netherlands; Hansen, B.G., European Chemicals Bureau, Ispra, Italy. Previous applications (Langenberg et al., SAR QSAR Environ. Res. 5:1-16, 1996) of the BIODEG Program to predict MITI (Ministry of International Trade and Industry, Japan)-I biodegradation test data have not been very successful. This may be due to microbial toxicity at the high concentration of test substance (100 mg /L) specified in the protocol; substantial differences in the degradative activity of the microbial population in the MITI-I test in comparison to other tests used for developing the BIODEG model; or other factors. To investigate this and possibly develop a new predictive model for ready biodegradability in the MITI-I test, a dataset of approximately 800 chemicals with MITI-I data was divided into a training and validation sets. Using the training set, a new set of coefficients was developed by multiple regression vs. a suite of molecular fragments similar to those already in the BIODEG Program. The quality of predictions from the new model was then evaluated by comparing predicted and experimental results for the independent validation set. Reasons for differences in the two models will be discussed.

587 **The Negative Relationship between BSAF and K_{ow} for Extremely Hydrophobic PCBs.** Maruya, K.A.*, Lee, R.F., Skidaway Institute of Oceanography, Savannah, GA. Laboratory experiments that have shown the positive correlation between the biota-sediment accumulation factor (BSAF, a model parameter) and log K_{ow} (a predictor variable) for hydrophic organic compounds (HOCs) have been well-validated in the field. However, this has not been the case for extremely hydrophobic compounds, i.e. those with log $K_{ow} > 6$. In this study, we present field data which show a consistently negative correlation between BSAF and log K_{ow} for several species of estuarine biota exposed to saltmarsh sediments contaminated with Aroclor 1268, a technical mixture whose congeners average 8.5 chlorines per biphenyl. Differences in BSAF among species are attributed largely to their specific feeding habits. Within a given species, the degree of *ortho* chlorine substitution also affects the magnitude of BSAF and lipid-normalized trophic transfer factors. These results serve as field validation for laboratory studies that suggest a declining relationship between accumulation and hydrophobicity for highly chlorinated PCBs.

588 **Predicting the Aquatic Toxicity of Lubricants Based on Lubricant Composition.** Isola, D.*, Texaco, Beacon, NY; Giddings, J.M.*, Springborn Laboratories, Wareham, MA. Knowledge of product characteristics that determine the aquatic toxicity of lubricants makes it easier to design new products with favorable environmental profiles. We examined the chemical composition of 14 lubricants, determined or estimated the aquatic toxicity and oil-water partition coefficients of product components, and then predicted the chemical composition and aquatic toxicity of Water Accommodated Fractions (WAFs) of each lubricant. The analysis identified components responsible for the toxicity of certain products to fish, invertebrates, and algae. The calculations accounted for most, but not all, of the differences in WAF toxicity among products and differences in sensitivity among species. Predictions were quantitatively accurate when data were available for nearly all potentially toxic components. For moderately toxic products, lack of toxicity data on components resulted in underestimation of WAF toxicity. As the utility of this approach for product stewardship becomes clear and the benefits are manifested as increased product acceptance in the marketplace, it should become easier to justify the effort to obtain the necessary data on the components.

589 **Response-surface Analyses for Toxicity to *Tetrahymena pyriformis*: Reactive Carbonyl-containing Aliphatic Chemicals.** Cronin M.T.D. and Schultz T.W.* School of Pharmacy and Chemistry, Liverpool John Moores University, Liverpool, England and College of Veterinary Medicine The University of Tennessee, Knoxville, TN. A response-surface has been developed with *Tetrahymena pyriformis* population growth impairment toxicity data (log $1/IGC_{50}$), the 1-octanol/water partition coefficient (log K_{ow}), and the energy of the lowest unoccupied molecular orbital (E_{LUMO}). A statistically robust plane (log $1/IGC_{50} = 0.52$ (log K_{ow}) - 0.849 (E_{LUMO}) - 0.30, $n = 51$, $s = 0.32$, $r^2 = 0.830$, $F = 117$) was found for reactive carbonyl-containing chemicals. Chemicals included aldehydes acting by the Schiff-base forming mechanism of electrophilicity, a,b-unsaturated aldehydes and a,b-unsaturated ketones acting by the Michael-type acceptor mechanism of electrophilicity, as well as selected diones. Diones are thought to act by a variety of mechanisms. The a-diones act as selective binders to arginine residues, g-diones act as selective binders to tubulin, and b-diones act through an unknown mechanisms of action. Outliers to the above model fell into two groups: small reactive molecules (e.g., acrolein) that were more toxic than predicted and molecules where the reactive center was sterically hindered by an alkyl group (e.g., 2,4-dimethyl-2,6-heptadienal) that were less toxic than predicted.

590 **A Hierarchical QSAR Approach to Predicting Carcinogenicity of Chemicals.** Gute, B.D.; Grunwald, G.D.; Basak, S.C., Natural Resources Research Institute, University of Minnesota, Duluth, MN. A major objective of predictive toxicology is to estimate the carcinogenic potential of chemicals. Structural and functional criteria have been used by experts in assessing the carcinogenicity of molecules. We have developed a hierarchical quantitative structure-activity relationship (QSAR) approach utilizing increasingly complex parameters in estimating genotoxicity. Four classes of theoretical molecular descriptors; viz., topostructural, topochemical, geometrical and quantum chemical parameters; were used to classify a set of 113 chemicals as carcinogenic or non-carcinogenic. The addition of biological data from short-term tests, such as Ames' mutagenicity and Ito's liver bioassay, into the set of independent variables resulted in the creation of a model with increased accuracy in correctly classifying carcinogens. However, the addition of these biological endpoints did not increase the model's efficacy at classifying non-carcinogens. The percentage of correctly classified non-carcinogens was the same using only theoretical indices and combining these indices with biological endpoint data. The utility and limitations of this approach in assessing the carcinogenicity of chemicals will be discussed.

591 **Mercury Loading of Two Large Temperate Lakes in Northern Minnesota.** Persell, J.S.*, Smith, S.J., King, S.O., Minnesota Chippewa Tribe, Cass Lake, MN. Consecutive studies on two separate lakes were conducted to quantify mercury loading to each ecosystem and develop lake-specific fish consumption guides that take into account subsistence diets for the adjoining Bois Forte Indian Reservation (Lake Vermilion) and Leech Lake Indian Reservation. Lake Vermilion is a fixed-crest reservoir (surface = 29.6 km^2, average depth = 7.6 m, pH = 7.6, alkalinity = 22, color = 38) that has a rugged and complex shoreline. Leech Lake is a managed-crest reservoir (surface = 66 km^2, average depth = 3.9 m, pH = 8.2, alkalinity = 155, color = 6) located 137 km southwest of Lake Vermilion near the headwaters of the Mississippi River. Lake stations and tributaries were sampled monthly. Wet/dry deposition were sampled every two weeks. Fish of a variety of species and sizes were sampled for each lake. All samples were analyzed for total mercury using cold-vapor atomic fluorescence spectrophotometry (CVAFS). While the mercury concentrations in precipitation was almost identical for the two study periods, mercury concentrations were higher in Lake Vermilion than in Leech Lake for water (1.95 ng/L and 0.49 ng/L, average surface concentration) and for fish (1.19 $\mu g/g$ and 0.16 $\mu g/g$, for a 2 lb. walleye). The observed differences between the two lakes can be attributed to each lake's unique watershed characteristics and loading coefficients, water chemistry, and food web.

592 **Air/water Partitioning of Trifluralin: Effects of Melt Point and Other Factors.** Rice, C.P.*, Nochetta, C.N., U.S. Department of Agriculture, Beltsville, MD; Chernyak, S.M., U.S. Geological Survey, Ann Arbor, MI. Interfacial transfers of chemicals between air-water phases, described by Henry's law constants (HLCs), have a crucial role in the environmental distribution and fate of many pollutant chemicals and accurate determination of these values is important. Of the two phases, air and water, the aqueous properties are more impacted by environmental factors. Temperature plays a very important role on the degree of partitioning. For chemicals whose melt points lie in the range of typical environmental temperatures (among which are several pesticides), it would be expected that the transition from solid to liquid form (melting) of a chemical should affect it's dissolution properties. Trifluralin demonstrated this effect by exhibiting a shift in the slope of the line for measured HLC versus temperature from 5° Celsius to 70° Celsius. This shift occurred at 49° Celsius which is the melt point for trifluralin. The plots of HLC versus temperature for trifluralin in salt water versus distilled water had similar slopes but the HLCs for the salt solution were higher. This suggests a salting out effect on the solubility of trifluralin. The plot for HLC versus temperature in the salt water experiment was non-linear. There is no simple theoretical explanation for this.

593 **Short-term Fluctuations Within Long-term Atmospheric Declines of Pesticide Concentrations in the Great Lakes Region.** Cortes, D. R.* and Hites, R. A., School of Public and Environmental Affairs and Department of Chemistry, Indiana University, Bloomington, IN 47408. As a participant in the Integrated

Atmospheric Deposition Network, Indiana University measures atmospheric concentrations of pesticides at sampling sites near the shores of the Great Lakes. Samples are collected using high volume air samplers and acquired over a 24 hour sampling period. Particulate-bound pesticides are collected on a glass or quartz fiber filter, and gas-phase pesticides are collected on XAD-2 resin or polyurethane foam. Following solvent extraction and chromatographic clean-up, analysis is performed using a gas chromatograph with an electron capture detector. The sampling campaign began in 1990 and continues though the present day, with one sample acquired every 12 days. During a study of long-term temporal trends of pesticides at these sites, individual occurrences of elevated atmospheric concentrations followed by rapid decay were observed. While long-term atmospheric declines are on the order of years, these individual high concentration events are over within one to two months. These events occur at multiple sites, and for different pesticides. In some cases, correction for temperature effects is first necessary to see these events, and in other cases the events are apparent in the original data. We calculated and compared long-term and short-term declines of pesticides in the atmosphere using first order decay.

594 Stable Isotope Fractionation: a Potential Biogeochemical Tracer of Persistent Organic Pollutant Fate. Padma, T. V. * and Dickhut, R. M., VIMS, College of William and Mary, Gloucester Point, VA, USA. Many persistent organic pollutants (POPs) are susceptible to long-range atmospheric transport and can accumulate in regions spatially remote from the source. A POP's chemical properties affect its rate and extent of atmospheric transport. A volatile POP such as hexachlorocyclohexane (HCH) is likely to migrate long distances prior to condensation at a sufficiently cool temperature. Chlordane, which is less volatile than HCH, should migrate at slower rates. Similarly, stable isotopes of a chemical are likely to migrate at different velocities since the heavier isotope will be less volatile. We tested these hypotheses using laboratory simulations of atmospheric transport and air-water exchange along a temperature gradient. Three interconnected air-tight gas washing bottles, containing 500ml seawater each, were placed in three respective water baths maintained at different temperatures (40°C, 20°C and 0°C). The inlet of the 40°C bottle was connected to an air source and the outlet of the 0°C bottle was connected to a tenax trap in order to collect any gas-phase organic contaminants present in overflow from the last bottle. The 40°C bottle was spiked with a perdeuterated and non-deuterated POP for each simulation. Stable isotope ratios for the POPs were monitored daily by subsampling the water in each bottle. Tenax traps were monitored every alternate day. POP concentrations were determined using gas chromatography and mass spectrometry. Results suggest that geochemical tracers such as stable isotope ratios may ultimately be useful tools to aid in evaluating the extent of long versus short-range atmospheric transport of POPs.

595 Temporal and Spatial Trends of Gas Phase Polychlorinated Biphenyls in the Great Lakes Atmosphere. Matt F. Simcik and Ronald A. Hites, School of Public and Environmental Affairs and Department of Chemistry, Indiana University, Bloomington, IN 47405. The Integrated Atmospheric Deposition Network (IADN) was conceived and implemented by the United States and Canadian governments to assess the role of atmospheric deposition of toxic contaminants to the Great Lakes. The network consists of one master atmospheric sampling station on each of the five lakes removed from the influence of local sources and several satellite stations, some of which are near urban areas. The three American master stations for Lakes Superior, Michigan, and Erie have produced data from their inception to August, 1997. All sampling equipment is located within 1 km of the shoreline. Sampling was performed every 12 days for 24 hours using high-volume air samplers equipped with a quartz fiber filter and either polyurethane foam plug or Amberlite XAD-2 resin as the adsorbent. Gas phase polychlorinated biphenyls (PCBs) at the three American master stations were strongly dependent on ambient temperature leading to the calculation of enthalpies of volatilization. Several differences were observed between the enthalpies for samples of over-water air masses and those of over-land air masses. All enthalpies were much lower than predicted from laboratory studies and Henry's Law calculations. Lake Superior showed no decreasing trend with time for PCBs either over-water or over-land. Lakes Michigan and Erie had significant decreases in gas phase concentrations over the course of IADN sampling. Half-lives associated with the Lake Erie over-water and over-land samples and the Lake Michigan over-land samples were similar ranging from 2.5 to 3.5 years. The Lake Michigan over-water samples indicated that there was a significant volatilization event in November 1992 after which the gas-phase total-PCB concentration decreased rapidly with a half live of about 130 days.

596 Hexachlorobenzene in Atmospheric Samples from Five Canadian Locations. Waite, D. T.*, Environment Canada, Regina, SK, Canada; Thompson, T. S. Saskatchewan Health, Laboratory and Disease Control Services Branch. Regina, SK, Canada; Gurprasad, N. P., Environment Canada, Environmental Protection Branch Laboratory, Edmonton, AB, Canada; Cessna, A. J., Agriculture and Agri-Food Canada, Saskatoon Research Centre, Saskatoon, SK, Canada; Quiring, D. W., Environment Canada, Regina, SK, Canada. Hexachlorobenzene (HCB), frequently reported as a contaminant in atmospheric samples from around the world, has been used as an agricultural fungicide and also occurs as a contaminant of some pesticides and industrial chemicals and a product of waste incineration. HCB was detected in most of the atmospheric samples collected from three Canadian locations in the summer of 1994 and seasonally, at five locations, from the summer of 1995 to the spring of 1996. Concentrations ranged from <0.005 to 0.112 ng m^{-3}. Median values from the various locations ranged from 0.008 to 0.011 ng m^{-3} for 1994 and 0.021 to 0.047 ng m^{-3} for the 1995-96 samples. At four of the sites in 1995-96, there was a slight, positive correlation between increasing air temperatures and increasing concentrations of HCB. At the fifth site, the correlation was negative with HCB concentrations decreasing with increasing air temperatures. Although HCB is a potential contaminant of pentachlorophenol (PCP), a wood preservative, there was no correlation between PCP concentrations and HCB concentrations in the same samples.

597 The Influence of the Gas Phase on Semi-Volatile Organic Contaminant Concentrations Found on Aquatic Particles. Jeremiason, J.D.*; Stapleton, H.M.; Baker, J.E., Chesapeake Biological Laboratory, Solomons, MD; Eisenreich, S.J., Rutgers University, New Brunswick, NJ. The gas phase influences semi-volatile organic contaminant (SOC) concentrations on aquatic particles through the combination of air-water exchange and solid-water partitioning. In theory, SOC concentrations on aquatic particles can be predicted based on the gas phase concentration, Henry's Law, and a solid-water equilibrium partition coefficient. In real systems, temporal changes in the gas phase concentration, production of particles by photosynthesis, settling of particles, etc. prevent air-water-particle equilibrium from being achieved. In particular, the incorporation of SOCs into phytoplankton and subsequent settling has the potential to lower dissolved SOC concentrations in surface waters creating non-equilibrium conditions favoring *net* air to water exchange. In this manner, the air-water-particle linkage has the potential to incorporate "new" loadings of SOCs to the base of aquatic food webs. The objective of this paper was to examine SOC air-water-particle relationships for contaminants including polychlorinated biphenyls (PCBs) and polycyclic aromatic hydrocarbons (PAHs) in Lake Superior, Grand Traverse Bay (Lake Michigan), and two small lakes on the Canadian Shield to determine the potential for "new" atmospheric SOC inputs. In all systems sediment traps were utilized to quantify vertical SOC settling fluxes. The magnitude of SOC settling fluxes in all systems has the potential to lower dissolved SOC concentrations in the surface waters. In the Canadian Shield lakes, PAH settling fluxes and *net* air to water exchange rates were similar in magnitude demonstrating the intimate linkage between the gas phase and SOC concentrations on aquatic particles. In the Great Lakes systems, settling fluxes were usually much greater than air-water exchange fluxes demonstrating intense internal recycling of SOCs.

598 Pesticides in the Air, Rain and Surface Water of a Chesapeake Bay Watershed. McConnell, L.L.*, USDA, Agricultural Research Service, Beltsville, MD; Nochetto, C.B., University of Maryland, Chesapeake Biological Laboratory; Rice, P.J., USDA, Agricultural Research Service, Beltsville, MD. An extensive study of air, rain and surface water concentrations of twenty-five currently used pesticides, organochlorine pesticides and three degradation products was conducted during the Summer of 1996 in the Patuxent River watershed. Air and rain were collected at three locations and water was collected at four representative locations in different areas of the river. Pesticide concentrations in water from an earlier study in 1995 combined with usage information from the Patuxent watershed indicate that a significant source of pesticides is located in the upper watershed area in Prince George's and Anne Arundel counties. Pesticide concentrations were compared with river flow and estimated use patterns in the watershed. Atrazine, 6-amino-2-chloro-4-isopropylamino-*s*-triazine (CIAT), simazine, metolachlor, and chlorpyrifos were consistently detected with maximum concentrations of 3000, 800, 2700, 70, and 190 ng/L, respectively. Concentrations were highest in the upper watershed, an area where 70% of pesticide usage occurs, and decreased with distance down the river. Dilution appears to be the main factor leading to the decrease in atrazine concentrations down the river. The expanded study conducted in 1996 will be used to more carefully determine the spatial and temporal variability in pesticide fluxes from runoff and atmospheric deposition to the entire watershed area and to describe the effects of tidal dilution on the fate of pesticides in this Chesapeake Bay tributary.

600 A Continuous Stirred Tank Reactor Exposure System for Investigating the Role of Vegetation in the Fate of Semi-Volatile Air Pollutants. Maddalena, R.L.*; N.Y. Kado, University of California, Davis, CA 95616; T.E. McKone, Lawrence Berkeley National Laboratory and University of California, Berkeley, CA 94720. Recent work suggests that terrestrial plants can influence the fate of environmental contaminants. In addition, the transfer of these contaminants from the environment to vegetation can introduce hazardous chemicals into the food chain. To understand the role of plants in the multimedia system, the factors that control the rate and extent of chemical partitioning between vegetation and adjacent environmental media need to be measured. Exposure chambers are useful for isolating and investigating individual transfer pathways through which environmental contaminants interact with vegetation. An exposure system based on the principle of a continuous stirred tank reactor (CSTR) has been described and used extensively for exposing plants to volatile air contaminants. The CSTR exposure system is adapted here and characterized for use in investigating the transfer of semi-volatile air contaminants to above ground vegetation. The system provides a controlled environment for exposing the above ground parts of mature plants to both ambient air pollutants and to contaminants introduced into the atmosphere using a gas phase generator column. By changing air contaminant levels and simultaneously monitoring the time dependent contaminant concentration in both the air phase and the tissue of the exposed plant, data on the rate and extent of chemical partitioning is obtained. The partitioning and mass transfer characteristics of phenanthrene, anthracene, fluoranthene and pyrene are estimated by fitting measurements collected during the exposure of mature *Capsicum annuum* (bell pepper) plants to the mathematical representation of mass balance in the air/plant system.

601 Swainson's Hawk Monitoring in the Argentine Pampas. Goldstein, M.I. and Lacher, T.E., Jr., Texas A&M University, College Station, TX; Sarasola, J.H., Universidad de La Pampa, Santa Rosa, La Pampa, Argentina. Swainson's hawks were monitored from November 1996 through February 1997 in the Argentine agricultural grasslands. Two sites were chosen, the first in northern La Pampa and the second, approximately 500 km away, in northeastern Cordoba. Due to extensive hawk mortality in 1996, the organophosphate insecticide monocrotophos (MCP) was removed from La Pampa markets prior to the 1996-97 season. It remained available, however, for use in Cordoba. Swainson's hawks were live-trapped in La Pampa (132) and Cordoba (14). Hawk condition was assessed, blood taken for cholinesterase (ChE) activity analyses, and ethanol footwash rinses and feather clippings were taken for pesticide residue analyses. Hawk ChE activities from apparently normal birds at two sites in North America were not significantly different from each other nor from South American samples. No chemical residues were found on birds from either site. An additional hawk from each province was brought in by farmers for rehabilitation. Both had inhibited ChE activities. Farmers in La Pampa were surveyed with questionnaires to determine chemical use strategies. No reported uses of monocrotophos occurred nor were mortalities found in La Pampa. One incident of 24 dead hawks was found in Cordoba. Carcasses were scavenged and desiccated and no chemical residues were found on remaining wing feather samples. Testimonies of surrounding farmers were inconclusive, although they used MCP illegally during this time for grasshopper control. Changing chemical use practices, as well as a more conducive climate (wetter, better crop growth, less agricultural spraying), all interacted to reduce mortality in the Pampas.

602 Pesticide Impacts on Swainson's Hawks and the Development of a National Ecotoxicological Monitoring Program in Argentina. Zaccagnini, M.E., Instituto Nacional de Tecnologica Agropecuaria (INTA), Parana, Entre Rios, Argentina; Mustacciolo, A., Servicio Nacional de Sanidad y Calidad Agroalimentaria (SENASA), Buenos Aires, Argentina; Lichtschein, V., Secretaria de Recursos Naturales y Desarrollo Sustentable (SRNyDS), Buenos Aires, Argentina, Canavelli, S., Uhart, M., INTA, Parana, Entre Rios, Argentina; Salto, C., INTA, Rafaela, Santa Fe, Argentina; Colazo R., INTA, La Pampa, Argentina; Gonzalez C., INTA, Oliveros, Santa Fe, Argentina. The Swainson's hawk (SWHA) is a neotropical migrant that spends northern summers in western North America and austral summers on Argentina's Pampas region. 6,000 SWHAs died on the Pampas from 1995 to 1996 due primarily to exposure to the organophosphorus insecticide, monocrotophos, which was used to control insect infestations. Argentine and US researchers were joined by academic, government, non-government and chemical industry representatives from Argentina, U.S., Switzerland and Canada in an international effort to avert recurrence of the deaths. Monocrotophos use restrictions, its withdrawal in the hawk's range, ecological studies on hawk movements on agricultural landscapes and outreach through extensionist, television, radio, and popular press information campaigns were initiated. Training programs prepared field and laboratory personnel for further incidents. These actions and favorable weather conditions held hawk mortalities to 24 in 1997 and 800 in 1998. This presentation details incidents, responses and their effectiveness and the proposal for initiating a National Ecotoxicological Monitoring Program in Argentina. The goal of this program is to control chemical impacts on wildlife, both migratory and local. Field, laboratory and administrative program components will be combined to provide the capability to detect ecotoxicological problems and focus resources on controlling their causes. Working cooperatively between government, academic and non-government organizations will optimize the resources available to, and the effectiveness of, the program.

603 Conflicts Between Dickcissels and Venezuelan Agriculture. Basili, G.*, Florida Audubon Society, Winter Park, FL; Temple, S.A., University of Wisconsin, Madison, WI. Dickcissels (Spiza americana) are a neotropical migratory songbird. On their breeding grounds in North America, they are appreciated for their beauty, song and insectivorous diet. On their winter range in Latin America, Dickcissels feed on rice and sorghum, and they are a serious pest to agriculture. Consequently, there exist contradictory attitudes and actions concerning Dickcissel conservation and management. On their temperate breeding grounds, efforts are being made to enhance Dickcissel populations. At the same time, there is movement to reduce Dickcissel numbers in tropical wintering areas. Although most farmers on their central wintering grounds in Venezuela use non-lethal controls to keep Dickcissels out of their fields, some resort to the use of toxic agricultural chemicals to intentionally kill Dickcissels. Herein lies the most immediate conservation concern. Dickcissels are extremely gregarious during winter, and a single roost can contain up to three, possibly 30% of the entire species population--and targeting such a megaroost could devastate a large portion the world's Dickcissel population. Chemical control and the resulting heavy mortality during the non-breeding season was likely responsible for the species' precipitous decline in the 1960's and 1970's. Although the population has recently stabilized, lethal control of Dickcissels continues in Venezuela. In addition, farmers continue to suffer economic hardships as a result of Dickcissels depredating their fields. In this paper, we review the conflict between Dickcissels and Venezuelan farmers, and we examine international actions underway that share responsibility for the stewardship of this migratory species.

604 Risk Characterization of Canadian Neotropical Migrants Exposed to Pesticides in Latin America. Baril A., National Wildlife Research Centre, Canadian Wildlife Service, 100 Gamelin blvd., Hull, Québec, K1A 0H3, Canada. In the context of increasing harmonization of regulatory requirements and trade across North and South America there is a need to expand pesticide risk assessment on a similar scale. Canadian bird populations are put at risk by the use of toxic pesticides both in Canada during the breeding season and outside of Canada on their wintering grounds. In this project we characterize the risk to bird populations from pesticide use on their wintering grounds. The following information was collected: extent of southern migration, nature of wintering habitat, feeding guilds, bird use of crops on the wintering grounds, pesticide use by crop type, and crop distributions in Latin America. This information is combined using a geographic information system for the purpose of screening the risks posed to each species. Over 190 species of neotropical migrants breeding in Canada winter in Latin America, of which 88 are known to associate with agricultural habitats. Despite numerous data gaps risks are characterized by crops. Levels of risk are also examined based on ecological considerations. The results are also discussed with respect to their relevance to resident species, the regulatory and risk management options at an hemispheric scale, and the possibilities for international collaboration on technology transfer, risk reduction, research and monitoring.

605 The Need for Global Restrictions of Problem Pesticides to Protect Migratory Birds. There are some pesticides whose toxicity to birds is so high that most agricultural use patterns with those compounds result in predictable, repeated and, largely unavoidable, mortality when birds are present. The way to deal with those products is to restrict allowable use patterns to those where exposure potential is negligible or, if this is impractical, to ban them altogether. Yet, regulatory actions taken in a single country are inadequate at protecting migratory birds. A clear example of this has been the problems experienced with the mortality of Swainson's Hawks (*Buteo swainsoni*) on their wintering grounds in Argentina. The insecticide monocrotophos was responsible for those kills. This insecticide had been restricted in the U.S., in part as a result of the extreme hazard to birds, but continues to be used extensively in the Americas in a wide variety of crops. A recent review of the field record reveals that bird mortality had been frequent and generally extensive in a wide variety of crops worldwide, a problem that should also have been easy to predict based on a risk assessment of available toxicity data. The case of granular formulations of carbofuran, is similar. Repeated and unavoidable mortality resulting from the attractiveness of the silica granule base as well as the extreme toxicity of the product to birds has been demonstrated in a wide variety of use patterns. Regulatory action was taken in Canada and the United States but the use of the product continues unabated in most countries where our birds winter. A mechanism is needed to achieve effective use restrictions of problem pesticides throughout the range of migratory bird species.

606 Mangrove Mutations Associated with Mercury Deposited in Molted Feathers of Scarlet Ibises. Temple, S.A.*, University of Wisconsin, Madison, WI; Klekowski, E.J., University of Massachusetts, Amherst, MA; Siung-Chang, A.M., Pan American health Organization, Port of Spain, Trinidad and Tobago; Kurmarsingh, Institute of Marine Affairs, Chaguaramas, Trinidad and Tobago. An unusually high incidence of mutations in a local population of red mangroves on the island of Trinidad was associated with locally elevated mercury levels in the underlying sediments. This restricted mercury contamination was spatially correlated with a large roost of scarlet ibises. Molted feathers of ibises using the roost had high concentrations of mercury, which were probably acquired by the birds during their annual migration to wetlands in South America contaminated with run-off from gold mining operations. Decades of molted feathers accumulating around the roost apparently has created a local contamination problem.

607 Accumulation of Persistent Organic Pollutants in the Eggs of Southern Ocean Migratory Albatross. Jones, P.D.*, ESR (Institute of Environmental Science and Research), Lower Hutt, NEW ZEALAND; Robertson, C.J.R., Department of Conservation, Wellington, NEW ZEALAND., Kannan, K., Giesy J.P., Department of Zoology, National Food Safety and Toxicology Center and Institute of Environmental Research, Michigan State University, East Lansing, MI. There is currently concern about the levels of persistent organic pollutants (POPs) accumulating in wildlife from locations previously thought to be "pristine". This is particularly the case for north pacific albatrosses which have been recently shown to carry considerable body burdens of a range of POPs. It is likely that these contaminants represent the general level of contamination of the north pacific ocean rather than specific point sources. Due to this global contamination we have conducted comparable analyses on albatross species from the south pacific ocean. The species studied, the Northern Royal Albatross (*Diomedea sanfordi*), Northern Buller's Mollymawk (*Diomedea* (*platei*) (nova sp.)) and Chatham Island Mollymawk (*Diomedea eremita*) have different migratory ranges covering most of the southern ocean. Egg samples were analysed for a range of POPs including polychlorinated- dibenzo-*p*-dioxins dibenzofurans and biphenyls as well as a range of organochlorine pesticides. Contaminant concentrations were considerably lower than in northern hemisphere albatrosses with international toxic equivalents (I-TEQ) concentrations ranging from 3.03 to 6.32 pg/g wet weight and total PCB concentrations ranging from 18.0 to 66.7 ng/g wet weight. One sub-group of samples showed significant egg shell thinning however this does not appear to be related to contaminant burdens. A risk assessment based on contaminant concentrations suggests that as for the northern pacific albatrosses I-TEQ and PCBs pose the greatest threats to southern albatross, however greater margins of safety are evident in the southern ocean birds.

608 Seasonal Changes in Contaminant Levels in Nearctic Shorebirds: Effects of Migration. Noble, D.G., University of Cambridge, Cambridge, U.K.; Braune, B.M.*, Canadian Wildlife Service, Hull, Quebec, Canada. We measured concentrations of contaminants in tissues of four Nearctic shorebird species, collected during five stages of their annual cycle (breeding, fall migration, early and late overwintering in Venezuela, and spring migration). Mean levels were below those associated with toxic effects. Semipalmated plovers tended to be most contaminated, short-billed dowitchers and semipalmated sandpipers had intermediate residues, and lesser yellowlegs were least contaminated, but significant interspecific differences were found only for DDT compounds, total PCBs and the higher-chlorinated PCBs, and renal cadmium. Significant effects of the stage of migration were found for DDT compounds, dieldrin, mirex, HCH, chlordane compounds, total PCBs, tetra-chlorinated PCBs and liver mercury, but not HCB, heptachlor epoxide, higher-chlorinated PCBs, selenium, cadmium or lead. Highest levels of most compounds were found at Churchill, Manitoba, whereas dieldrin and DDD+DDT were highest at Delaware Bay, N.J. Concentrations of chlordane compounds and total PCBs were highest on arrival in Venezuela in the autumn, but these compounds, like other organochlorines, declined while overwintering in Latin America. Seasonal patterns suggest exposure to North American sources during migration as well as continued atmospheric deposition of volatile compounds to the Arctic. Interspecific differences in foraging behaviour and metabolism, other than the demands of egg-laying, appear to have less influence on variation among samples.

609 Organochlorine Pesticide Contamination in New World Passerines. Harper, R.G.*, Illinois Welseyan University, Bloomington, IL; Capparella, A.P., Illinois State University, Normal, IL; Klemens, J.A., Illinois Wesleyan University, Bloomington, IL; Frick, J.A., Illinois Wesleyan University, Bloomington, IL; Richardson, H.B., Illinois Wesleyan University. Few data exist on levels of organochlorine pesticide contamination in New World passerine birds (i.e. songbirds that reside in North America, Mexico, Central America, the West Indies, and/or South America). The purpose of our ongoing research is to document the extent and patterns of organochlorine contamination in Neotropical migrant passerines (those that breed in North America north of Mexico, and winter in Mexico, Central America, the West Indies, and/or South America), and in resident North and South American passerines. The most frequently detected compounds were p,p'-DDE, dieldrin, and heptachlor epoxide. Neotropical migrant passerines examined in our previous studies had a high frequency of contamination (85 of 93 individuals were contaminated, representing 17 species); residue levels ranged from 0.02 ng/g to 3540 ng/g. There were no significant differences in contamination levels between males and females, or between winter habitat type (forest or shrub). Surprisingly, South American resident passerines had a very low frequency of contamination (3 of 102 individuals collected in Argentina, Peru, and Guyana, representing 54 species), while many North American residents (21 of 27 individuals, representing 9 species) were contaminated at levels ranging from 0.4 ng/g to 67 ng/g. These results suggest that Neotropical migrant passerines are acquiring organochlorine contaminants from their North American breeding grounds, their non-South American wintering grounds, or as they migrate through Mexico and Central America.

610 Patterns of Bioaccumulation of PCBs in Nestling Tree Swallows on the Hudson River, NY. Echols, K.R.*,Tillitt, D.E., Meadows, J.C. Gale, R.W., Peterman, P.H. Environmental and Contaminants Research Center, USGS-BRD, Columbia MO; Secord, A., McCarthy, J. US FWS, Cortland, NY. Tree Swallows (*Tachycineta bicolor*) were used as sentinel species for monitoring PCB uptake along the Hudson River. Three sites and a reference site were established as tree swallow colonies in 1994 and 1995 around and downstream from Fort Edward. Collected samples in 1995 were tree swallow eggs, adult females and chicks at days 5, 10 and 15. Additionally, insects and insect mouth samples were taken for analysis of food source PCBs. All samples were analyzed for 102 PCBs and combined PCB congener peaks including mono-*ortho* and non-*ortho* substituted PCBs. Eggs from contaminated sites ranged from 9 μg/g to 25 μg/g of total PCBs. By day 15 chicks had accumulated total PCBs from 24 μg/g to 96 μg/g at the contaminated sites. Adults had concentrations ranging from 21 μg/g to 114 μg/g. Reference levels were 4.5, 1.4 and 22 μg/g for eggs, chicks and adults, respectively. Accumulation rates in chicks at the contaminated sites caused masking of growth dilution effects. PCB congener concentrations will be modeled according to work by Nichols et al. (1995, ES&T, 29, 604-612) Using principal components analysis, and pattern recognition, eggs, chick and adult tree swallows were evaluated for PCB bioaccumulation and metabolism and compared to patterns from insects and sediment.

611 The Use of Epidemiology in Risk Assessment. Rebecca L. Calderon, U.S. EPA, RTP, NC. Epidemiology has been considered the fundamental science of public health policy. The use of epidemiologic data in environmental health policy has been limited particularly in the environmental regulatory arena. Epidemiologic risk assessment is different from risk assessment and the interplay between the two has lead to some misconceptions over the use of epidemiologic data. The current risk assessment process was designed in a time when the need for regulation was great and the epidemiologic information was sparse. There was little time for the consideration of conducting specific studies to improve the information base for environmental health policy. Animal bioassays could be conducted under standardized protocols within defined time periods. The limitations and uncertainties of animal studies also became standardized and risk assessors became comfortable with their models of extrapolation. As the cost of regulations have grown, the economic realities of regulating with little or no data to support actual public health benefit have become a political and legal liability. Epidemiology has a much to promise and in actual practice is used more widely than believed. EPA has regulated several chemicals in which the key regulatory information used was epidemiology. In many environmental media, environmental epidemiology is becoming less and less of a black box. Several major scientific advances in exposure assessment and human toxicology have increased the ability of scientists to conduct high quality epidemiology. Since human data is the "gold standard", it is anticipated that the need for epidemiology will increase as the Agency moves forward into the 21st century.

612 An Objective Analysis of the Environmental and Human Health Risks Associated with Pharmaceuticals. Webb, S.F., Procter & Gamble Technical Centres Ltd., Staines, UK. Environmental assessments are required for new pharmaceuticals in both the USA and EU. Despite this, previous discussions regarding the nature and form of risks to the environment and human health from pharmaceuticals have taken place in the general absence of a systematic analysis of their potential impacts. This paper attempts to address this deficiency via an objective retrospective review of existing ecotoxicity data and a consideration of potential environmental and human exposure. Ecotoxicity data relating to >100 human pharmaceuticals (excluding estrogens) have been collated. They suggest a lack of acute effects at concentrations <100 µg/l in fish, algae or invertebrates. The availability of UK usage data (1995) permitted risk characterisation (i.e., PEC/PNEC calculation) for >60 compounds. PNEC values were derived via application of an assessment factor of 1000 to acute endpoints (i.e., L(E)C_{50} values). Under "*worse-case*" exposure assumptions of no human metabolism, passage of all material to drain, no removal during wastewater treatment and no surface water dilution of effluent, the large majority of pharmaceuticals considered yielded PEC/PNEC ratios of <1 suggesting no concern in the aquatic environment. For the remainder, incorporation of a consideration of typical surface water dilution (DF = 10) and realistically expected removal during wastewater treatment (in the case of Acetaminophen) was sufficient to yield PEC/PNEC ratios <1. Calculation of potential life-time ingestion via drinking water employing "*worse-case*" assumptions (as above and with no removal during drinking water treatment) revealed I_{70} values (based on the ingestion of 2 litres/day for 70 years) equivalent to ≤2 days of the corresponding daily therapeutic doses. The only exceptions were Aspirin and Acetaminophen where the I_{70} values were equivalent to <10 days of the daily therapeutic doses.

613 The Role of Ecological Epidemiology in Regulatory Risk Assessment. Veith, G.D., US EPA, Research Triangle Park, NC The role of ecological epidemiology in regulatory risk assessment is poorly defined due, in part, to the ambiguities in terminology and a greater preference for evidence from controlled experiments. EPA is making a major effort to refine epidemiological methods to address the large-scale assessment questions with respect to natural resources as part of the Environmental Monitoring and Assessment Program (EMAP). This researchprogram is developing methods for measuring the health of ecosystems as well as monitoing designs to spatially describe the distribution of health status in the environment. Weak associations are formed in EMAP through measures of stressors such as toxic chemicals, hypoxia, nutrient and habitat quality. These findings have been used to identify hazards at a regional scale and shape the priorities for regulatory programs. Moreover, EMAP has a special focus to understand the dynamic exchange between epidemiological methods and experimental research at our laboratories and process research in small ecosystems. This presentation will discuss some of the EMAP findings from freshwater and estuarine ecosystems in the Mid-Atlantic region.

614 Hemochromatosis: Interactions Between Genetic Disease and Environmental Hazards. Preston, B.L.*, School of Biology, Georgia Institute of Technology, Atlanta, Georgia; McDonnell S.M., Dietz W.H., National Center for Chronic Disease Prevention and Health Promotion, Centers for Disease Control and Prevention, Atlanta, Georgia; Buchanan S.D., National Center for Environmental Health, Centers for Disease Control and Prevention, Atlanta, Georgia. Hemochromatosis (HC) is a genetic disease of iron regulation, affecting 1:200-1:400 in the U.S. One of 10 persons is a carrier, making HC the most common known genetic disorder. Pathogenesis is characterized by increases in intestinal absorption and hepatic retention of iron. The resulting chronic iron toxicity promotes a variety of chronic diseases including liver cirrhosis, cardiomyopathy, and diabetes. Because the absorption of other divalent cations and liver dysfunction may be increased in HC, individuals with HC may be more sensitive to a variety of environmental hazards. We reviewed literature (1965-present) from the U.S. Environmental Protection Agency, Natural Resource Defense Council, Centers for Disease Control and Prevention, Medline, and environmental databases for environmental hazards which may potentially interact with HC. Persons with HC may be at increased risk for adverse responses to certain environmental hazards, associated with drinking water, food supplies, recreational areas, and occupation. The increased absorption and retention of iron associated with HC has been correlated with concomitant increases in the absorption and/or retention of other metals including lead, aluminum, cobalt, copper, zinc, and manganese. The liver damage commonly associated with HC may increase the probability of adverse responses to these and other toxic challenges due to decreased capacity for hepatic detoxification. In addition, liver dysfunction combined with high blood concentrations of iron may increase the susceptibility of individuals with HC to certain types of bacterial and viral infections. Environmental hazards are an important public health concern for this sensitive subpopulation. Further research and case detection should be conducted to describe fully the size of this subpopulation and characterize the risks associated with environmental hazards. Finally, the communication of these risks to individuals with HC, health care providers, and environmental regulatory agencies may reduce the likelihood of adverse effects associated with HC.

615 Integrating Health Surveillance and Environmental Monitoring. Epstein, P.R. The emergence of new diseases, and resurgence and redistribution of old diseases in the past two decades is one symptom of a more generalized environmental distress syndrome. While multiple factors contribute to emerging infectious diseases (EIDs), ecological dynamics and biodiversity losses can amplify climatic factors underlying EID emergence. Genetic, species, population, functional group and habitat diversity are all important in providing ecosystem resilience and resistance to invasion of exotics and infestations; and predator/prey relationships are key in controlling the proliferation of pests and pathogens. Three key groups (rodents, mosquitoes, plankton) — opportunistic organisms that increase their populations rapidly in response to environmental disturbance — may serve as biological indicators of alterations in ecosystem vulnerability. Monitoring their abundance, species composition and distribution can help integrate health surveillance with environmental monitoring systems and standardize monitoring at such as EPA sites, NOAA Large Marine Ecosystem (LME) Programs, and Long-Term Ecological Research Sites (LTER - NSF funded). Data from the various disciplines must be "fused" and combined into advance Geographic Information Systems to map consequences, causes and costs. Such integration is now occurring between the Centers for Disease Control and Prevention and the Sevilleta LTER in the US Southwest, with respect to rodent populations and the hantavirus pulmonary syndrome. There are several implications of such integration: 1. Can we better asses the impacts of global change? 2. Can we improve our understanding of the environmental causes and costs of EIDs through investigations of linkages among physical and biological earth systems? And 3. Can Health Early Warning Systems be developed based on climate forecasts combined with RS/GIS of conditions conducive to epidemics, for implementation of timely, environmentally-sound public health interventions?

616 Hantavirus Outbreaks and Rodent Ecology: The Role of El Niño. T. L. Yates1*, T. Ksiazek[2], R. Parmenter[1], J. Mills[2], P. Rollin[2], S. Nichols[2], J. Dunnum[1], R. J. Baker[3], C. Parmenter[1], J. Childs[2], and C. J. Peters[2]. [1]Department of Biology and Museum of Southwestern Biology, University of New Mexico, Albuquerque, NM, [2]Center for Disease Control and Prevention, Atlanta, GA, [3]Department of Biology, Texas Tech University, Lubbock, TX. *Hantavirus* is a genus of the family Bunyaviridae that is closely associated with wild rodents. In 1993, a newly discovered hantavirus caused an outbreak of human disease in the Southwestern U.S. with a mortality rate of over 50%. The cause of the outbreak and the basic biology of the virus and its host were not known. To better understand these factors, rodent population densities and levels of hantavirus infection were examined across six different habitats at the University of New Mexico's Long-term Ecological Research Site in central New Mexico and at four longitudinal sampling locations throughout the state. Rodent samples were tested from habitats ranging from Pinion-Juniper Woodland to Desert Grassland beginning in 1989. Additional samples archived in frozen tissue collections also were examined from throughout the range of the reservoir species dating to 1979. In addition, evolutionary trees of hantaviruses and their rodent hosts were compared in order to gain an understanding of the co-evolution of this rodent-virus association. Seroprevalence in populations of *Peromyscus* varied as a function of habitat and population density. At low density the number of habitat types harboring rodents infected with *Hantavirus* was reduced relative to the number found with infected populations at times of increased density. These data were found to be strongly correlated with global climate changes and fluctuations including the El Niño Southern Oscillation (ENSO). Comparisons between virus and rodent family trees reveal a long and close evolutionary association. These data coupled with those from studies of archived samples suggest that new world *Hantavirus* are very old and probably arrived in North America over 20 million years ago. Outbreaks appear to be positively associated with rodent population densities which in turn are correlated with climate change and human land use patterns.

617 "Health" in "Environment": a Canadian Perspective. Bickis, U.I.*, Phoenix OHC, Inc., Kingston, ON, Canada; Kwiatkowski, R. and David, C., Health Canada, Ottawa, ON, Canada. The objective of this presentation is to demonstrate how information, knowledge and expertise from human health specialists are being integrated into the environmental assessments of projects, by an environmental community that has traditionally been largely preoccupied with the assessment of impacts on ecosystem components other than the human health aspect. Following a brief description of Canadian regulatory infrastructure, roles and responsibilities, the presentation will outline methodologies, initiatives and case studies involving the incorporation of ecosystem and human health considerations into impact prediction and risk assessments. These are drawn from the authors' experiences, not only in Canada, but internationally. The presentation provides an overview of perspectives on, and approaches taken towards, the integration of human health and ecosystem health into a holistic environmental framework. The insights gained should be useful for attendees interested in developing a more integral approach to their practice.

618 Screening Level Risk Assessment for Open Water Disposal of Dredged Material: Case Study of Pilgrim Nuclear Power Plant (Plymouth, MA, USA) Intake Channel Sediments Containing ^{60}Co. Lombardo, V., US EPA Region I, Boston, MA; Kullberg, P.G., US Army Corps of Engineers, New England District, Concord, MA; Tomey, D., US EPA Region I, Boston, MA. Radioactive wastes and other matter are prohibited from ocean disposal in greater than *de minimis* amounts. This applies to low-, mid-, and high-level radioactive wastes and other materials. EPA Region I conducted an initial assessment of the environmental significance of measured levels of ^{60}Co in sediments to be dredged from the intake channel of the Pilgrim Nuclear Power Plant and proposed for disposal at the Massachusetts Bay Disposal Site (MBDS). Specifically. a screening level risk assessment (RA) calculation was performed to evaluate the potential human health effects from consumption of fish which feed at the MBDS (conservative, upper bound estimate) following disposal of dredged sediment. The calculation showed that disposal of the material with the measured low (trace) level of ^{60}Co would have no adverse effects on human health. Briefly, the RA calculation showed that in order to receive even a "trivial dose" of radiation from the sediments (0.01 mSv/yr, according to the International Atomic Energy Agency) an adult human would have to consume 62 lbs. of "contaminated" fish (feeding exclusively at the disposal site) every day of the year. Based on that finding, it was also predicted that exposure to the measured ^{60}Co level would not have any unacceptable effects on marine animals because the scientific literature indicates that marine animals are less sensitive to radioactivity than humans. Subsequent results of toxicity and bioaccumulation testing supported the screening level RA.

619 Ecological and Human Health Risks at an Outdoor Shooting Range. Peddicord, R. K., Dick Peddicord & Company, Inc., Parkton, MD. Ecological and human health risks from shooting-related contaminants were quantified in association with rifle/pistol and trap/skeet shooting in wetlands and wooded areas. Contaminants of concern included lead, arsenic, antimony, copper, zinc, and PAH associated with shotgun targets. These were examined in sediment, soil, surface water, ground water, air, and tissues of birds and wildlife. Receptors of concern included shooters, range workers, trespassers, deer, fox, grouse, robins, phoebes, rabbits, mice and hawks. No contaminant risks specific to the site were identified for shooters. Range workers could face slight risks from inhalation of dust and incidental ingestion of soil or sediment. Trespassers illegally poaching and consuming deer on range property could be subject the same elevated risks from arsenic as hunters consuming deer taken throughout the surrounding area due to background arsenic levels. Child trespassers playing on active ranges after shooting hours could face slight risks from lead in incidentally ingested soil, sediment and water. Direct consumption of lead shot by grit-ingesting birds and by small mammals such as rabbits and mice could pose risks to those individual organisms consuming the shot. Species that might directly ingest shot tend to be highly fecund and have populations over contiguous areas much larger than the range, so that only a small proportion of the breeding population might ever be exposed to the range. Population density-dependent compensation mechanisms would likely offset any contaminant-related loss of individual organisms. There could be some risks to individual predators of organisms that directly ingest shot. Individual predators feed over a much larger area than the range. The proportion of their diet that might contain shooting-related contaminants is sufficiently small to result in minimal risks to individual predators and even less risk to predator populations. No ecological risks were predicted at ecosystem, community or population levels.

620 Human Health Risk Assessment of Emissions from Hazardous Waste Combustion Facility Using Updated U.S. EPA 1998 Protocol. Morton, E.S.*, Tetra Tech EM Inc., Chicago, IL; Yurk, J.J., U.S. EPA, Dallas, TX; Desmond, W.P., Tetra Tech EM Inc., Dallas, TX; Matthews, B.M., Tetra Tech EM Inc., Dallas, TX; Riggins, C.R., Tetra Tech EM Inc., Dallas, TX; McDonald, T.B., Tetra Tech EM Inc., Dallas, TX; Woo, C.I., Tetra Tech EM Inc., Dallas, TX; Pryor, A.G., Tetra Tech EM Inc., Dallas, TX. The 1998 U.S. EPA Office of Solid Waste multipathway human health risk assessment protocol is described showing its application for performing a human health risk assessment for ISCS3T modeled stack and fugitive emissions from a hazardous waste incinerator. Methods for COPC identification, site characterization, fate and transport assessment of emissions, identification of exposure scenarios and locations, and pathway risk calculations are described showing the major steps in identifying pathways of concern. Updated procedures for evaluating breast milk exposure and acute inhalation exposure are presented. Updated procedures for assessing mercury risk, including environmental speciation and food chain transport are also presented. Application of the protocol methods to determine carcinogenic risk and noncarcinogenic hazard of COPCs is described for a site in EPA Region 6. Default exposure parameters were initially used to identify pathways of concern. The potential risk of these pathways was reassessed with additional site-specific meteorological, land use, and exposure fate and transport information. Results were used to identify issues of concern to address in the RCRA permitting process. The use of stand-alone multipathway risk assessment software is described showing the simplification of the risk assessment process.

621 Ecological Risk Management - Past, Present and Future. Huggett, R. J.* Michigan State University, East Lansing, MI; Oppelt, E. T., U.S. EPA, Cincinnati, OH. The term, "ecological risk management," is relatively new in our vocabulary. However, humans have been employing ecological risk management, in one form or another, for thousands of years. Biblical writings tell or suggest ways to eliminate noxious pests and pestilence by leading a more "righteous" life. Protecting fish or game populations by setting aside spawning areas and habitat, by establishing seasons, and bag or creel limits are examples of efforts that started to gain momentum only in the last century or so. Controlling the concentrations of toxic substances in surface waters through the use of water quality standards and criteria go back only a few decades. Sediment quality criteria, green chemistry and life cycle or "cradle-to-grave" analyses are even more contemporary attempts at ecological risk management. Risk management at the community or ecosystem level of biological organization is now upon us and one could argue that recent international efforts to stem CFCs and control carbon dioxide emissions are attempts at ecological risk management at the global level. As the level of sophistication relative to ecological risk management has increased, the necessity to involve stakeholders has become more apparent. Economic, political, social, spiritual, statutory or legal aspects of management options need to be considered. In fact, science or engineering may not play the paramount role in choosing a particular management option. In the future, we will likely see the trend of ecological risk management focused on higher levels of biological organization continuing. A better understanding of effects of complex mixtures will lead to regulations aimed at managing suites of compounds. Biomarkers will likely play a increased role in determining management end points and managing ecological risks that have an international component will be more common. Finally, we expect climate change to be a major factor in ecological risk assessments and management decisions.

622 Environmental Risk Management: The Road Less Traveled. Biddinger, G.R.*, Brown, R.P., Durda, J., Gala, W.R., Harrass, M.C, Pittinger, C.A. and Stahl, R.G., Members of the American Industrial Health Council's Ecological Risk Assessment Committee, Washington, DC. The U.S. Congress in the 1990 Clean Air Act Amendments mandated a Presidential/Congressional Commission on Risk Assessment and Risk Management (i.e., the President's Commission). The final report of the President's Commission (1997) provided a clear statement of "Principles for Risk Management Decision-Making". These principles provide broad guidance to risk managers, relevant to both human health and ecological risk. The American Industrial Health Council (AIHC) Ecological Risk Assessment Committee building upon the work of the Multi-stakeholder Ecological Risk Management Dialogue Group (MERMD), and the results of the SETAC technical workshop on ecological risk management (Williamsburg, VA, June 1997) applied the President's Commission's principles to specific ecological applications. A series of practical criteria were identified for gauging whether and how the President's Commission's principles were followed for particular ecological risk scenarios. These criteria will be presented in the context of organizational management activities (e.g., planning, design, implementation, etc.) and specific types of applications. Barriers to the effective integration of these principles by the regulated community will be highlighted and recommendations for improvement are provided. Lastly, factors that affect the relative importance of attributes to success and the criteria upon which that success is evaluated will be discussed.

623 Integrated Environmental Decisionmaking. Matanoski, G.*, Johns Hopkins University, Baltimore, MD; Miller, T. and Sanzone, U.S. EPA, Washington, DC. The Science Advisory Board (SAB) has completed a three-year study, involving more than fifty scientists, into improved approaches for incorporating scientific information (including economics and some other social sciences) into an integrated process for making environmental decisions. Building upon, but extending beyond, its 1990 Reducing Risk report, the SAB report provides general guidance and specific tools for distinguishing among competing environmental risks and among risk reduction options. While encouraging the EPA's more goal-driven agenda and the involvement of interested and affected parties throughout the process, the SAB recommends additional steps that the Agency can take to systematize the application and evaluate the results of the decisionmaking process. The proposed "integration" includes a) technical integration (using information on multiple-stressors, multiple-pathways, multiple-endpoints, etc.); b) comparative integration (comparing risks posed by different stressors); c) management options integration (considering the entire suite of risks that are addressed by each management option); d) economic integration (considering cost/benefit and cost/effectiveness issues); e) feedback integration (evaluating the effectiveness of the adopted risk reduction action and making necessary adjustments); and f) values integration (reflecting public values throughout the process).

624 Valuing Environmental Impacts: Lessons Learned from the Natural Resource Damage Debate. Cantor, R.*, LECG, Inc., Washington, DC. In environmental regulations, achieving a balance across competing or confounding objectives is the norm rather that the exception. This insight could not be more true than in the areas of ecological risk management and assessing natural resource damages, where trustees and responsible parties are faced with the daunting task of meeting objectives that include making the public whole, minimizing transactions costs, and facilitating timely remediation and restoration activities. Recent federal natural resource damage rules, however, convey a distinct preference for resource-to-resource and service-to-service considerations for restoration project selection and scaling, and only after those approaches prove inappropriate to trustees does economic valuation become a basis for planning. This paper addresses why ecologists and economists disagreed in their support for these rules. It reviews the criticisms raised by the environmental constituency about the general model of economic valuation and its assumptions. Much of the ecology/economics debate centers on distinctions between efficiency and equity, particularly intergenerational equity. There are, however, more subtle and long-standing causes for the divide that stem from fundamentally different concepts of value and its measurement. The paper concludes by considering the potential of net environmental benefit analysis to mitigate ecology/economics controversies in managing ecological risks and natural resource planning.

625 Integrated Ecological Risk Management: Considering Remediation and Restoration Options. Strong, A. L.* and Campbell, T. A., Campbell & George, L.L.P., Houston, Texas. Until recently, the risk assessment/risk management process was largely focused on the protection of human health. Risks to ecological receptors were rarely considered. Today, however, both human health and ecological risks are given equal consideration. As a result, agencies are now faced with making ecological risk management decisions at sites where ecological risks are determined to exist. EPA and several states have addressed this issue by stating that the site risk manager must balance risk reductions associated with cleanup of contaminants with the potential impacts of the closure/remedial actions themselves. While this is a sound approach, achieving this balance can be extremely difficult in the absence of a scientific decision-making framework. This is attributable, in part, to the complexities associated with ecological risk assessments which focus on the effects of multiple contaminants to numerous potential receptors such as birds, mammals, and fish, all having different exposure tolerances, body burdens, etc. Presently, these regulatory agencies are developing regulations, guidelines and policy regarding assessing and managing ecological risks. In undertaking these efforts, we have urged that these agencies adopt a *substantive* ecological risk management framework that results in full integration of remediation and restoration options and seeks to balances the risks and the benefits of potential options to the environment (*i.e.* one that evaluates the ecological risks present at a site in relation to the overall ecological services provided by that site's habitat). The presentation will provide examples of the application of this integrated approach for several major remediation sites in the Gulf Coast region where wide-spread support for the selected option came from federal and state regulatory agencies, the public, environmental groups, and the responsible parties. Integration of remediation and restoration-based alternatives into the decision-making process results in "common sense" approach to ecological risk management decision-making. It is technically-defensible, protective of the environment, and cost-effective.

626 Managing Towards Sustainability: An Environmental Framework for Business. Feijtel, T.C.J.*, Hindle, P., Hill, J.A., de Saedeler, D.J., Saouter, E., The Procter & Gamble Company, Brussels Belgium; White, P.R.; Newcastle England; Pittinger, C.A., Cowan, C.E., Cincinnati Ohio. Sustainable Development (Sustainability) is a broad public policy concept lacking clear definition, thus hindering effective business action in managing towards sustainability. This barrier must be overcome, as technological progress towards sustainability clearly implies innovation, and business is the engine of innovation in the marketplace. Without clear guidance on what "moving towards sustainability" means in practical terms, business has no focus against which to manage. The objective of managing towards sustainability can be defined as, "Environmentally (and technically) feasible, socially acceptable environmental decision-making". From this admittedly broad definition, it is possible to develop an environmental framework of principles and management tools to assess: safety; regulatory compliance; resource use and waste management; and the addressing of societal concerns. In each of these four areas a business objective can be defined against which decisions can be tested. Such decisions are based on technical, economic and social data and arrived at within a decision-making organisational structure. Such an approach provides a practical basis upon which a company can manage its day-to-day business and improve continuously its sustainability performance. This environmental management framework is practically illustrated for consumer products by the incorporation of key assessment tools satisfying each of the four elements.

627 Ecological Risk Management. Champagne, L.F., TNRCC, Austin, TX. Ecological risk management (ERM) can be defined as a process which balances site risk reductions obtained through remedial actions with environmental, legal, economical, social, and technological considerations; whereas, ecological risk assessment (ERA) has been defined as a process that evaluates adverse ecological effects as a result of exposure to stressors. Some believe that these two processes are separate and ordered and should remain so. They contend that the risk assessor need only focus on the site-specific ecological issues and report the ERA results to the risk manager who will then make an autonomous decision on the remedy. Others feel that the two processes must be intertwined in order to make well-informed decisions. For instance, although ERM at a corrective action site is the ultimate responsibility of the risk managers, it is essential that communication with the risk assessors and stakeholders occur throughout the process in order to achieve a "balanced" decision. Similarly, risk managers can utilize their "big picture" perspective to help risk assessors set meaningful assessment endpoints during the ERA. It is not important that the risk managers understand all the intricacies of the ERA. However, they should at least be familiar with its assumptions, resolutions, and particularly its uncertainties, as these are critical to evaluating the overall appropriateness of any remedy. When evaluating these remedial options, risk managers should consider several types of information in addition to the ecologically-protective cleanup levels derived from the ERA. This information should include: short- and long-term effectiveness of the remedy, natural remediation (i.e., no action), public acceptance, cost, implementability, and potential remediation impacts. A somewhat controversial risk management option which has emerged recently is the consideration of compensatory restoration (i.e., creating or setting aside usually like habitat to offset residual contamination). However, in order for this to be a truly viable option, the Natural Resource Trustees should be involved, as they are the experts in identifying and negotiating "environmental currency" and ensuring public acceptance. This option brings risk management to a new level - one where remedial action and natural resource damage liability issues come together. In these cases, the challenge for the risk manager will be to work closely with the Trustees in much the same way as he/she works with the risk assessors. For these and other reasons, risk management should be viewed as a multifaceted process which is neither linear nor unilateral.

628 *Pfiesteria* **Hysteria in the Chesapeake Bay Ecosystem During the Summer of 1997.** deFur, P.L. and C. Fehl. Virginia Commonwealth Univ., Richmond Va. In the summer of 1997, the dinoflagellate *Pfiesteria piscidida*, was found in fish in the Pokomoke River, MD. The Pokomoke River forms the border between Maryland and Virginia on the Eastern Shore, the Delmarva Peninsula, an area where fishing, agriculture and chicken farming are dominant practices. During the fall of 1997, a group of faculty and graduate students from the Center for Environmental Studies at VCU assessed four aspects of the then-current discovery of the *Pfiesteria* infestation: ecological considerations, including *Pfiesteria* life-cycle; human health effects; economic consequences; and legal issues. It was clear that only part of the ecological picture is understood. The life cycle of *Pfiesteria* is known in part only, and the environmental conditions that promote or favor its growth and proliferation are unclear. Run-off from confined livestock feeding operations was implicated with the *Pfiesteria* outbreak, but the connections and cause and effect relations have not been well quantified or described. Human health effects were thought to be limited to high exposure cases in confined spaces, but since then, Maryland state agencies reported cases of human effects from ambient exposures at the river site. The economic consequences were reportedly severe, but not well quantified in terms of lost sales or altered use habits. Seafood sales did seem to decrease and recreational use of several water bodies declined sharply. The legal avenues for controlling the apparent causal factors (e.g. run-off or nutrient addition) are weak, at best. Land-use controls, water quality permits and non-point source run-off control programs could offer some means of addressing the problem. The two states of Maryland and Virginia took fundamentally different approaches to policy decisions for this complex case with limited scientific information.

629 **Linking Ecological Risk Assessment to Human Health Risk Assessment: Food-chain Transfers to Humans from Subsistence Ecological Resources.** Himmelbauer, L.D.*, Alaska Department of Environmental Conservation, Juneau, AK. Bartell S., Haness S., Nobmann E., Ponce R., IDM Consulting, Seattle, WA. Many of the contaminated sites in Alaska are in rural or remote locations that may overlap ecological resource areas partially used by Alaska Native Americans and others for subsistence hunting, fishing, and gathering. Fish consumption data for Native American subsistence populations are very limited, and no data had currently existed that quantified intake rates for non-fish subsistence resources such as caribou, moose, mussels, etc. Sets of subsistence ingestion exposure factors were developed for various regions in Alaska to use to determine food-chain transfers to humans from contaminated ecological resources. Five EcoRegions that reflect the predominant Alaska Native culture associated with the various major ecological regions were selected for analysis: Arctic-Subarctic, Aleutian Pacific, Subarctic Interior, SE Alaska Coast, and Urban. Exposure intake rates (mean, 50th percentile, 95th percentile) for these ecoregions were developed through statistical analysis (Monte Carlo) of the harvest and consumption data of more than 200 villages in Alaska. The five highest harvested resources in Alaska were determined to be 1) salmon; 2) non-salmon fish; 3) large land mammals, such as caribou, moose, bears; 4) marine mammals, such as seals, otters, whales; and, 5) marine invertebrates, such as mussels, clams, and crab. The subsistence intake rates will be used as an exposure assessment tool to identify top subsistence food resources consumed in a study area; to identify data needs when performing site-specific risk assessments; to calculate preliminary risk estimations for communities when contaminant information is available; and to calculate Reasonable Maximum Exposures (RME).

630 **Interacting with Regulatory Agencies and Stakeholders and Site-specific and Regional Risk Assessments.** Menzie*, C.A., Menzie-Cura & Associates, Chelmsford, MA; van der Schalie, W.H., U.S. EPA, National Center for Environmental Assessment, Washington, D.C. Over the past several years the U.S. Environmental Protection Agency (USEPA) has been developing guidelines for the conduct of ecological risk assessment (USEPA, 1992; 1997). During the course of this process, the agency has become increasingly aware of the role that stakeholders have in the process. This has become evident through the agency's experience with the conduct of assessments, from comments submitted to the agency, and from the emphasis given to the role of stakeholders in recent reports by the Commission on Risk Assessment and Risk Management (1996) and the National research Council (NRC, 1996). This paper identifies "lessons learned" related to involving stakeholders in the ecological risk assessment process. Specifically, it is based on experience gained during the Problem Formulation phase of risk assessments that have addressed chemical, physical and biological stressors at various spatial scales. These include assessments of ecological risks associated with hazardous waste sites (local scales), watershed programs (local to regional scales), and shrimp viruses (local to regional scales). These programs differ in purpose and complexity. Our paper will describe common features of stakeholder involvement as applied to these diverse cases. We will also identify how regulatory and stakeholder involvement varies among ecological risk assessment applications.

AA01 Ethics in Ecological Risk Assessment: An Overview. Maciorowski, A.F.*, US EPA, Washington, DC; Reinert, K., Rohm and Hass Company, Spring House PA; Rodgers, J., Jr., Clemson University, The Institute of Wildlife and Ecological Toxicology, Pendeleton, SC. Ecological risk assessments (ERA) may be conducted for a variety of reasons. Some ERAs are conducted for purely scientific purposes and may be viewed within an objective, value-neutral context. Other ERAs are specifically conducted for application in a regulatory decision making process that includes scientific, as well as social, economic, political, legal, and policy-based components. In this broader arena, objective value-neutral scientific positions are important, but are also subject to substantial challenge. Scientific and legal challenges may arise from opposing interpretations of the same data, differing disciplinary perspectives (e.g., agronomy, chemistry, ecology), as well as differing interpretations of regulatory policy and precedents by different public interest groups. Such challenges may range from collegial to highly adversarial, and may result in litigation. Given the complex subject matter and often contested application of risk assessments in the regulatory arena, it is somewhat surprising that a formal dialogue of ethics in ecological risk assessment has not yet surfaced. This paper presents an overview of ethical issues in risk assessment to initiate such a dialogue.

AA02 The Normative Function of Ecosystems in the Era of Environmentalism. Krimsky, S*. Department of Urban & Environmental Policy, Tufts University, Medford, MA. This presentation will explores the biological concept of the ecosystem as a fundamental source of value in environmentalism. Terms like *ecosystem health*, *biodiversity*, *ecology-based policy* and *ecological economics* are examples of how ecosystems are serving transcientific roles in linking science to environmental ethics and prescriptive policy making. The advantages and limitations of building a system of environmental values from the concept of *ecosystem* are discussed and linked to ecological risk assessment.

AA03 Ethics in Ecological Risk Assessment. Moore, D.R.J.*, The Cadmus Group, Ottawa, ON. The use of ecological risk assessment in environmental decision making has had a checkered ethical history. The reasons for this are due more to our misguided beliefs concerning public involvement in risk assessment than to overtly unethical or deceitful practices. The first misguided belief is that public involvement in the decision making process is unnecessary or restricted to the role of mere receptacles for downloading information. Public involvement is required for several reasons: (i) they have a crucial role in deciding what the focus of the assessment should be – it may not always be the most sensitive species that matters most, (ii) the public should decide how conservative assessors and managers ought to be – the decision to use hyperconservative safety factors is not one that should be made by assessors in isolation, and (iii) the public needs to understand the assessment approach and methods in order to contribute to the decision making process in a meaningful way. Recently, Environment Canada, the US EPA and others have recognized that public involvement is a crucial aspect of the decision making process and have begun acting accordingly. The second misguided belief is that assessors ought not to confuse the public by stating our uncertainties and expressing risks in a probabilistic form. Rather, we generally express risk as a precise number (quotient = 3.24) or effects as certain or uncertain. Such preciseness is not warranted on appeal to the evidence, and the public knows it. If our objective is to produce credible risk assessments, then we must engage the public early and often, and provide them with full information regarding risks. Full information means explicitly stating what we know and what we do not.

AA04 Ethics in Ecological Risk Assessments Involving Agriculture. Shafer, S.R.*, US Dept. of Agriculture, Office of Risk Assessment and Cost-Benefit Analysis, Washington, DC. Agriculture, risk assessment, and ethical frameworks all embody ideological positions (i.e., viewpoints of what is, what is considered *good*, and what can be). A framework for ethics acknowledges the degree to which a social group extends rights to some other. Agricultural systems are managed to maximize the reliability of producing food and fiber for human use and would not exist without human intervention and need. Some ecological characteristics considered typical or desirable in natural ecosystems are not compatible with agricultural production. Agricultural systems are embedded within larger landscapes, and the incompatibilities between optimal characteristics of the natural and managed systems lead to risks posed by each system to the other. In a risk assessment that incorporates information about agricultural systems, the system (or stakeholders for the system) selected for preferential rights by the risk assessor probably will be readily apparent through statements about which system (agricultural or natural) is considered the source of stressors, which processes or substances are considered undesirable, and which system contains the resources considered at risk. Even when the agricultural system is considered the source of stressors to natural systems, the requirement of society for the relevant agricultural products should be acknowledged in the assessment. Wise management of ecological risks is highly desirable to conserve or preserve natural resources, however, risk management options that reflect an environmental ethic should be examined carefully for risks posed to systems that provide for fundamental human needs.

AA05 Ethical Treatment of Environmental Issues by Business. Biddinger, G.R., Exxon Company, U.S.A., Houston TX; Reinert, K.R., Rohm & Haas Company, Spring House, PA.; Pittinger, C.A., The Procter & Gamble Company, Cincinnati OH; Stahl, Jr., R.G., E.I. Du Pont de Nemours & Co. The belief that the private sector and those who work in it are ethically challenged is ill founded. Some environmental activist organizations are prone to imply that the concept of *business ethics* is an oxymoron. This is a joke that is easy to make and for an uniformed listener just as easily accepted. The purpose of this paper will be to raise the level of awareness among SETAC members with regard to how industry ethically manages the environmental aspects of its products and operations. Approaches used by business to assure that their employees are performing their day-to-day activities in an ethical fashion are reviewed. How environmental risk assessment principles and methodologies are addressed through typical management systems, controls, policies, and training practices is presented. Additionally, examples of how, if current *Command and control* approaches were to evolve to provide for flexible management solutions, industry could exceed current environmental performance.

AA06 Ethics and Ecological Risk Assessment: A Regulatory Science Perspective. Maciorowski, A. F.*, Zeeman, M., US EPA, Washington DC. Ecological risk assessments for regulatory operations are science based in concept, historical development, and practice. However, they are applied in a complex, multi-dimensional decision-making arena that considers other natural and social science perspectives, as well as legal and political concerns. In this milieu, science, science policy, agency specific regulatory policy, and broader public policy concerns may be difficult to distinguish. This is particularly true when different public and private sector advocacy groups promote different values and belief systems as the only *right and proper* interpretation of data for particular situation or decision. Scientifically trained risk assessors are rarely adequately prepared for the dichotomous value systems, viewpoints, and belief systems they encounter in an inherently political regulatory process. Moreover, we are frequently asked how we are able to maintain personal and professional ethics and integrity in the regulatory arena. In this paper, we present examples where ecological risk assessments and values have collided and how they were resolved.

AA07 Environmental Decision Making: The Precautionary Principle or Risk Assessment? Raffensperger, C*., Science and Environmental Health Network, Windsor, ND. The precautionary principle is an environmental decision-making tool which stands in contrast to risk assessment. Science is essentially amoral, not immoral or moral. It is precisely this context which provides the (un)ethical dimension. As Maya Angelou says, fact is not truth and facts can obscure the truth. Risk assessment has been criticized by the environmental community because it is inherently undemocratic, does not acknowledge the larger societal context or give deference to the vast uncertainties inherent in, particularly, ecological risk assessment. Whether risk assessment is invoked by GATT, NAFTA, or the Food Quality Protection Act, it has served private, rather than public interests. Risk assessment is used to bolster U.S. global economic competitiveness rather than used to protect the environment and public health. That is, its context has been unethical because it presumes that everything is for sale. Until recently, there have been no alternative decision-making tools offered which would better serve the public interest. The precautionary principle offers a rare opportunity to rethink the context and the questions underlying environmental decision-making. This paper will compare and contrast the ethical implications of ecological risk assessment and the precautionary principle and propose a method for making decisions which incorporate the rigor of risk assessment and the ethics of the precautionary principle.

AA08 Deus ex machina or scientific reasoning: Where do we go in educating our students in the science of environmental risk assessment? Solomon, KR, Centre for Toxicology, University of Guelph, Guelph, ON, Canada. This paper will contrast numerical approaches to the use of scientific reasoning in the educating university students in the procedures of risk assessment with the purpose of maintaining the highest standards of scientific ethics. Recognizing that it is impossible to know everything, the ethical focus in risk assessment is towards most closely approaching the truth in assessing the effects, fate and risks of physical, chemical and biological stressors in the environment. Numerical methods and procedures have been proposed for many of the steps in the risk assessment process. These procedures are less biased but they may not properly consider the role of organisms in the ecosystem. On the other hand, purely naturalistic approaches to risk assessment process. These procedures are less biased but they may not properly consider the role of organisms in the ecosystem. On the other hand, purely naturalistic approaches to risk assessment may err by assigning too much importance to organisms and their value to society. In educating students to conduct risk assessments to the highest ethical standards, how can we make sure that they are aware of the potential pitfalls in the process? How can we increase their knowledge of the scientific principle, the rules of casualty and consequence and the proper weighing of evidence? This presentation was briefly discuss basic principles, offer examples of correct and incorrect approaches, and suggest ways and means whereby we can improve our education practices to move studeats from awareness to ownership of high ethical standards in the science risk assessment.

AA09 Summary and Questions to the Author Panel. Maciorowski, A.F.*, US EPA, Washington, DC; Reinert, K., Rohm and Hass Company, Spring House PA; Rodgers, J., Jr., Clemson University, The Institute of Wildlife and Ecological Toxicology, Pendeleton, SC. The final paper in this session will provide a brief summary of the different ethical perspectives discussed and provide an opportunity for the audience to ask questions of the authors.

IP01 Relative Utility of Intrinsic and Extant Biodegradation Kinetics. Grady, C. P. L. Jr., Clemson University, Clemson, SC. The outcome of biodegradation testing for synthetic organic chemicals (SOCs) is influenced strongly by the physiological state of the biomass used in the testing protocol. At one extreme, when very small inocula are used with a sufficiently high substrate concentration, the bacteria attain unrestricted growth at their maximal rate. Kinetic parameters obtained from such tests are *intrinsic* and depend only on the structure of the SOC and the type of bacteria in the inoculum. At the other extreme, when very large inocula are used relative to the substrate concentration, the substrate is exhausted before significant new enzyme synthesis can occur. The resulting kinetic parameters are *extant* and represent the capability of the biomass in the environment from which they came; they are dependent on culture history as well as the SOC structure and the bacterial type. Studies were conducted with lab-scale activated sludge wastewater treatment systems to compare the relative utility of intrinsic and extant biodegradation kinetic parameters for predicting the fate of six SOCs supplied as part of a complex feed containing both synthetic and biogenic organic chemicals. Both types of kinetic parameters were measured with respirometry and used to predict the effluent concentrations of the SOCs. Although both types overpredicted effluent concentrations, predictions made with extant parameters were consistently closer to the measured values. In addition, the extant testing protocol was easily modified for considering the effects of simultaneous multiple substrate biodegradation on the observed biodegradation kinetics. This presentation will describe both intrinsic and extant testing protocols, present the data concerning the relative utility of the two, and discuss the implications of the findings to predicting the fate of SOCs in natural and engineered environments.

IP02 Microbial Growth Kinetics and Eco-physiology During Mixed-substrate / Multiple-nutrient-limited Growth: Implications for Pollutant Degradation in the Environment. Egli T., Swiss Federal Institute for Environmental Science and Technology, Duebendorf, Switzerland Physiological and kinetic studies on the degradation of pollutants in the laboratory typically use conditions where a pollutant is supplied as the only source of carbon and where a single known nutrient (e.g., carbon) is limiting growth. In contrast, degradation of pollutants in the environment takes place usually in the presence of a multiplicity of alternative growth substrates of natural origin and it is often not known which nutrient limits growth. Therefore, it remains to be established whether and how the information obtained from pure culture / single substrate (pollutant) studies in the laboratory can be applied to ecosystems. Recently, laboratory studies using pure cultures and defined substrate/nutrient mixtures have provided considerable evidence that under environmental conditions microbial cells degrade pollutants together with natural (carbon) substrates and that growth can indeed be limited by two or more nutrients at the same time. Selected examples will be presented which demonstrate the effect of the simultaneous consumption of mixtures of substrates on growth and degradation kinetics and how the observed kinetics may be linked to the expression of pollutant degrading enzymes. Furthermore, a concept will be presented which allows for prediction of the range where microbial growth is at the same time limited by two or more nutrients. The consequences of multiple-nutrient-limited growth conditions on the degradation of pollutants will be discussed.

IP03 Advances in Environmental Microbiology and Molecular Biology and Their Application to Improved Biodegradability Testing and Environmental Exposure Assessment. Snape, J.R.* & Evans, M.R. Brixham Environmental Laboratory, ZENECA Limited, Freshwater Quarry, Brixham, South Devon, TQ5 8BA, UK. Biodegradation is a key removal process controlling the fate, concentration and distribution of chemical compounds in the environment. Consequently, removal due to biodegradative processes is used, together with other data, to determine exposure concentrations in environmental compartments. Within the European Union, the pass/ fail outcome of a standardised ready biodegradation test (OECD 301 series) is used to assign arbitrary first order rate constants for removal in waste water treatment plants, surface waters, soil and sediment. Recent attention has focused on replacing these default rate constants with measured rate constants, through the use of kinetic models based on those of Monod. As a result of this attention, improvements have been made to the existing biodegradation test methods that increase both their data quality and predictive nature. Automated respirometers measuring oxygen uptake and/or carbon dioxide evolution are rapidly emerging as the next generation test methods. Whilst improving the predictive nature of these screening tests is important, the question of the environmental relevance is equally, if not more, important. The issue of environmental relevance must be satisfied before biodegradation rates, measured in the laboratory, can be applied to the environment. This presentation will explore and identify approaches to addressing the question of environmental relevance within the context of biodegradation testing. Particular attention will be paid to the use of molecular techniques as part a multi-disciplinary approach to solving some of the historical problems associated with environmental microbiology.

IP04 Quantification of biodegradation test results obtained at low and high (screening level methods) concentration of test substance. M. G. Christensen and N. Nyholm, Department of Environmental Science and Engineering, Technical University of Denmark. A series of manometric BOD biodegradation screening tests were carried out in OECD mineral medium with a selection of model compounds and inocula. derived from activated sludge, river water and coastal water, respectively. With a sufficiently low inoculated biomass (e.g. 3 mg/L of activated sludge) fairly reproducible estimates of the maximum specific growth rate of the degrading consortium of micro-organisms were obtained when negative test results were discarded. Rates were independent of both source, pre-treatment (freshly collected or pre-exposed) and concentration of the inoculum, except that degradation was slower in seawater than in freshwater. Average growth rates were as follows: aniline (AN) 1.6 d^{-1}; 4-nitrophenol (4NP) 1.5 d^{-1}; 2,4-D 0.9 d^{-1}, Maleinhydrazide : 0.2 d^{-1} ; 4-chloroaniline (4ClA): negative. Die away tests with the same chemicals and microbial matrices were conducted with test concentrations sufficiently low to ensure first order kinetics (5-25 µg/L). Both freshly collected and pre-adapted biomasses were used. Pre-adaptation was carried out using a semicontinuous procedure and applying the same concentrations as in the test. Again disregarding negative results, significant correlations could be established between growth rates from screening tests and first order degradation rates for either preexposed (0.12 - 0.22 d^{-1}) or non-exposed (0.09-0.33 d^{-1}) biomasses. While the relative degradability ranking of the model compounds was consistent from screening tests to low concentration tests, the environmental degradation rates could be predicted very approximatetly, only.

IP05 Removal of LAS from Three Yorkshire Rivers - Determination of removal rate for the GREAT-ER. (Geographically-referenced Environmental Assessment Tool for European Rivers) Project. Fox, K. K.* (Unilever, Port Sunlight, UK), Holt, M. (ECETOC, Brussels, BE), Daniel, M., (Environment Agency Laboratory Service, Leeds, UK), Buckland, H. (Yorkshire Water, Bradford, UK), and Guymer, I (University of Sheffield, UK). Linear alkylbenzene sulphonate (LAS) removal has been determined three Yorkshire rivers, of varying depth and containing different initial levels of sewage treatment works effluent. The rivers are Red Beck, a small Yorkshire stream which receives effluent from a periodically malfunctioning trickling filter plant scheduled for imminent replacement; the Maun, a relatively clean river which is approximately twice as deep, and the Aire downstream of Leeds, which with flows of about 10 cumecs is one of the most polluted rivers in Europe. For the smaller rivers a pulse of effluent from the treatment plant was marked with Rhodamine WT, and LAS and water quality parameters, including suspended solids and boron, were measured as the Rhodamine WT pulse passed downstream. Dilution due to increased baseflow in the catchment was determined from flow measurements as well as from boron and Rhodamine WT concentration data, with the Rhodamine WT data found to be less reliable, probably due to adsorption of the dye onto river sediments. For the river Aire, conductivity and flow measurements were used to follow the effluent pulse downstream, and LAS removal in an effluent packet was determined relative to the boron concentration. First order LAS removal half-lives, which ranged from 2 hours 40 minutes in Red Beck to over 30 hours in the Aire, are interpreted with respect to suspended solids concentration and settling behaviour, and to biodegradation. The results of this study will be incorporated in the GREAT-ER model for Tier 2 Risk Assessment.

IP06 Kinetic biodegradation tests at low concentrations with pre-adapted surface water. L. Toräng, F. Ingerslev and N. Nyholm, Department of Environmental Science and Engineering, Technical University of Denmark. A method is proposed for adaptation of surface water samples to environmentally realistic low concentrations of the test substance for subsequent use in kinetic biodegradation tests. Ordinary batch tests with freshly collected (unadapted) water and low chemical concentrations, often produce highly variable results. Rate constants can be extremely variable, and sometimes degradation may fail or stop after a certain time. Also with very long incubation times (several months) the water sample may deteriorate and the simulation value of the test decrease. To remedy these problems tests can be carried out with adapted water, but to obtain realistic rate constants *overadaptation* must be avoided and the exposure concentrations should therefore be as in the subsequent kinetic test. To meet these objectives simple semicontinuous adaptation procedure is suggested. The basic principle is to start the test as a normal batch test and then periodically renew one third of the water with freshly collected site water every 1-2 weeks making up test substance to the starting concentration As the procedure comprises both a re-inoculation and a compensation of any depleted primary substrates the initial microbial diversity and substrate availability are restored and the feasible duration of the batch tests thereby extended to infinity in principle. Once adaptation has taken place, the semicontinuous operation is simply discontinued and the degradation test conducted as a usual batch test. The degradation rate resulting can be interpreted as the rate characteristic of an adapted system. Practical results demonstrating the feasibility of the procedure are presented with aniline (reference compound), 4-chloroaniline and 2,4-D in stream water.

IP07 The Non-equivalence of Pass Criteria in Standardised Ready Biodegradation Tests. Snape, J.R.* & Evans, M.R. Brixham Environmental Laboratory, ZENECA Limited, Freshwater Quarry, Brixham, South Devon, TQ5 8BA, UK. Biodegradation is a key removal process controlling the fate, concentration, and distribution of chemical compounds in the environment. Consequently, removal due to biodegradative processes is used, together with other data, to determine the impact a chemical substance will have on receiving environments. Within the European Union, data derived from standard biodegradation test methods are used in regulatory decision making to determine whether a chemical substance may pose long-term adverse effects to the aquatic environment and to evaluate environmental exposure concentrations. These regulatory decisions are based largely on the pass/ fail outcome of a simple biodegradation screening test, one of the six available ready biodegradability tests (OECD 301 series). The pass criteria for tests measuring respirometric activity is 60%, whereas tests measuring removal of dissolved organic carbon have a pass level of 70%. These pass criteria must be achieved within a 10 day time window. A recent research project, funded by the United Kingdom Department of the Environment investigating biodegradation test methods has demonstrated that these pass criteria are not equivalent. This was especially true at low test substance concentrations. All biodegradation studies were conducted using a radiolabelled version of the ISO CO_2 headspace test and a mass balance was obtained for all experiments. The non-equivalence of these criteria has serious regulatory implications regarding classification and labelling, and exposure assessment in environmental risk assessment. This presentation will provide data, for several test substances at a variety of concentrations, to demonstrate the non-equivalence of the ready biodegradation test pass criteria.

IP08 Investigation of an Onsite Wastewater Treatment System (OWTS) in a Sandy Soil: Part 4: Adsorption and Biodegradation of LAS Doi, J. and Marks, K., Roy F. Weston, Inc., Exton, PA; DeCarvalho, A., The Soap and Detergent Association, New York, NY; McAvoy, D., The Procter & Gamble Company, Cincinnati, OH; Nielsen, A.*, CONDEA Vista Company, Austin, TX; and Kravetz, L., Shell Chemical Company, Houston, TX. This work is part of an overall program for investigating the fate and transport of household cleaning product ingredients in onsite wastewater treatment system. The objective of this work was to determine sorption coefficient and biodegradation rate constants for incorporation into a predictive model for an OWTS. In both the sorption and biodegradation studies, three distinct soil samples were collected from a Jacksonville, FL septic tank field study site. Different concentrations of the test substance were added to a series of test vessels that contained upgradient groundwater and the collected soils. The adsorption test was designed to determine the partitioning of LAS between each soil type. Results indicated that the Freundlich isotherm coefficient (K) ranged from 0.55 -1.38, the adsorption coefficient (K_d) ranged from 0.43 - 4.02 and the rate of ultimate degradation (first order rate constant - K_1) ranged from 0.08 - 2.17 day^{-1} with increasing distance (0.3 - 1.2 m vertically and 0 - 6.1 m horizontally) from the leach field. All three soils showed that 60-80% of theoretical CO_2 was produced over the 30 day period of the test. These results demonstrate that both adsorption and biodegradation are key factors in the removal of LAS in a subsurface soil system.

IP09 Comparison of Measured Removal of LAS in Laboratory Activated Sludge Systems with Modeled Removal based on Batch Biodegradation Rates and Sorption Coefficients. Federle, T. W.*, Kaiser, S. K., McAvoy, D. C., Itrich, N. R., Gledhill, D. W., and Kerr. K. M. The Procter and Gamble Company, Cincinnati, OH. Continuous Activated Sludge (CAS) experiments were conducted with 1, 3 and 10 mg/L of added 14C(ring) C12 LAS to a to raw domestic sewage influent. At steady state, removal of parent and total radioactivity were measured. In addition, samples of mixed liquor were obtained from the CAS units and used for batch die-away and sorption experiments. Die-away experiments consisted of dosing the samples with a trace level (100 ug/L) of 14C(ring) C12 and monitoring the disappearance of parent, formation of metabolites, formation of 14CO2 and incorporation into solids with time. Sorption coefficients (Kd) were determined in a batch equilibrium experiment with inactivated sludge. First-order rates for primary degradation from the die-away tests were 32 - 41 hr-1 and appeared unrelated to influent concentration. Kd equaled 3500 L/kg. A wastewater treatment model, based on first-order biodegradation and equilibrium sorption processes and parameterized with these batch data and the CAS operating conditions predicted LAS removal of 99.2 - 99.3% with influent concentration having little effect on percent removal. Actual CAS measurements showed 99.3 - 99.4% LAS removal of LAS with influent concentration having little effect. These studies show that an activated sludge model parameterized with appropriate batch data can accurately predict removal in realistic conditions.

IP10 Biodegradation Kinetics of Nonylphenol and Nonylphenol Ethoxylates in Various Environments. Naylor, C.G.*, Huntsman Corporation, Austin, TX; J.B. Williams, Union Carbide Corporation, South Charleston, WV; J.E. Heinze, John Adams Associates, Washington, DC; C.A. Staples, Assessment Technologies, Fairfax, VA. NP and NPE biodegradation in aerobic laboratory and field systems has been extensively reported in the literature. Published studies include NPE and NP in standard bottle tests, NP in sea water, NP and NPE in soil, and NPE in activated sludge. Rates of degradation in most cases were not calculated from the data. Where the data are available, they generally follow first order kinetics, allowing calculation of rate constants and half-lives. Representative half-lives Iinclude 3 days for NPE mineralization in the SCAS test, 18 days in soil and 19 days in a river water die-away. For NP representative mineralization half-lives include 6 days in the OECD 301F test and in sea water with sediment, and 7 to 10 days in sludge-amended soil. Results from a comprehensive set of half-live values will be reviewed and critiqued. While laboratory biodegradation studies may not predict rates of degradation under real world conditions, they do provide weight of evidence for the biodegradation and non-persistence of NP and NPE in biological treatment and in the environment.

IP11 The Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC): Background and Overview. Benson, W. H.*, Environmental and Community Health Research Institute of Pharmaceutical Sciences, School of Pharmacy, University of Mississippi, University, MS. The Endocrine Disruptor Screening and Testing and Advisory Committee (EDSTAC) was chartered by the U.S. EPA to provide advice concerning the development of a strategy for screening and testing endocrine disruptor chemicals responsive to legislation enacted in 1996 (Food Quality Protection Act and Safe Drinking Water Act). The Committee consisted of approximately 40 scientists representing academia, industry, environmental and public health advocacy groups, labor, and small businesses. The Committee met eight times between December 1996 and June 1998. The committee formulated a series of recommendations that included: 1) a conceptual framework and related guiding principles; 2) a strategy for sorting and prioritizing chemicals for screening and testing; 3) a screening battery consisting of *in vitro* and *in vivo* assays; 4) a testing battery consisting of mammalian, avian, fish, amphibian, and invertebrate multi-generational studies; and 5) a communication and outreach strategy to inform the public of results of the screening and testing program. This presentation will provide background and an overview of the Committee process and early decisions regarding the need to consider: human and ecological effects; estrogen, androgen, and thyroid hormone related effects; and common mixtures.

IP12 The EDSTAC Conceptual Framework. Weis, J.*, Department of Biological Sciences, Rutgers University, Newark, NJ. EDSTAC established a conceptual framework to guide its deliberations and those of its subordinate workgroups. The Committee determined that a tiered approach would provide the most effective use of resources to detect endocrine disrupting chemicals and quantify their effects. The core elements of EDSTAC's recommended approach include: initial sorting; priority setting; Tier 1 Screening (T1S) and Tier 2 Testing (T2T); and hazard assessment. The Committee also provided principles to guide the overall development of the screening and testing strategy, decisions regarding the selection of screens and tests, how the T1S battery should be designed and used, and how the T2T battery should be designed and used. This presentation will provide an overview of the conceptual framework and guiding principles developed by EDSTAC to guide the remaining workgroups.

IP13 Priority Setting: Principles and Process. Osimitz, T. G.*, S.C. Johnson, Racine, WI; Walker, J., U.S. EPA, Washington, DC. The EDSTAC recommended sorting and prioritizing approximately 86,000 pesticides, commodity chemicals, naturally occurring estrogens, food additives, cosmetics, nutritional supplements, and representative mixtures for endocrine disruptor screening and testing. The sorting and prioritization approach relies on initial sorting of chemicals based on existing exposure and effects data and information. Next, high-throughput pre-screening (HTPS) using automated technology to perform transcriptional activation assays will be conducted on chemicals (approximately 15,000 chemicals produced in amounts greater than 10,000 pounds per year) with little or no effects data. The existing information and HTPS data will used to develop a relational database to synthesize and integrate data to develop a *compartment based approach* for further priority setting. Differn compartments would be based on different information categories such as: 1) integrated exposure and effects criteria; 2) exposure criteria only; 3) effects-related criteria only; and 4) specially targeted chemicals (nominations from the public, phytoestrogens, representative mixtures, etc.). The EDSTAC also recommended priority setting for Tier 2 Testing. This presentation will provide an overview of the priority setting process and principles recommended by EDSTAC.

IP14 The EDSTAC Screening and Testing Program. Borgert, C.*, Alachua, FL; Touart, L. EPA, Washington, DC. EDSTAC developed a two-tiered endocrine disruptor screening and testing program focused on estrogenic, androgenic, and thyroid effect endpoints. Tier 1 screening was designed to maximize sensitivity and minimize false negatives; include a range of organisms representing differences in metabolism; detect all known modes of action for the endocrine endpoints of concern; include a sufficient range of taxonomic groups among the test organisms; and incorporate sufficient diversity among the endpoints, permitting weight-of-evidence conclusions. The EDSTAC screening battery consisted of an estrogen receptor binding/reporter gene assay, an androgen receptor binding/reporter gene assay, a steroidogenesis assay with minced testes, a rodent 3-day uterotrophic assay, a rodent 20-day pubertal female assay with thyroid; a rodent 5 to 7 day Hershberger assay; a frog metamorphosis assay, and a fish gonadal recrudescence assay. Tier 2 testing was designed to evaluate the nature, likelihood, and dose-response relationship of chemicals. The EDSTAC testing battery consisted of a two-generation mammalian reproductive toxicity study or a less comprehensive test (e.g., alternative mammalian reproductive test), an avian reproduction test, a fish life cycle test, a mysid life cycle test, and an amphibian development and reproduction test. The committee also developed principles regarding weight-of-the evidence interpretation and validation needs for the individual screens and tests. This presentation will provide a detailed overview of the screening and testing batteries recommended by EDSTAC.

IP15 Interpreting Conventional Ecotoxicity Tests for Endocrine Disruption Effects. Touart, L.W. , U.S. EPA, Washington, DC. Recommendations from the Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC) for screening and testing chemicals for endocrine disrupting activity include new assays and modified versions of established assays. Conventional tests used to support pesticide registrations such as avian reproduction and fish life cycle tests are limited in discerning endocrine relevant endpoints. Interpreting the results of these tests for endocrine disruption requires understanding how the various endpoints relate to endocrine controlled processes While existing methods may be used to ascertain the apical relevance of an endocrine disrupting chemical, these methods are not sufficient to fully characterize the endocrine specific mode of action.. The utility of existing test methods and the value added by proposed modifications for risk assessment will be discussed.

IP16 EDSTAC Communication and Outreach: Improving Communication with the Public and Interested Parties. deFur, P.*, Center for Environmental Studies, Virginia Commonwealth University, Richmond, VA; Borgert, C., Alachua, Fl. The Endocrine Disruptor Screening and Testing Advisory Committee (EDSTAC) formed the Communications and Outreach workgroup to address issues concerning EDSTAC recommendations to the Environmental Protection Agency about the results of a screening and testing program once implemented. The workgroup addressed issues concerning: 1) how to best inform the public and interested parties about EDSTAC activities; 2) the best way to present the final outcome of the EDSTAC deliberations; and 3) how EPA might best communicate the results of the screening and testing program. The workgroup addressed the problem of accurately conveying the results of EDSTAC processes and the screening and testing program. The workgroup worked with EPA in setting up a database of parties interested in the outcome of the EDSTAC process and likely to be interested in the screening and testing program results. The outcome of the deliberations completed by EDSTAC concerning communication issues will be summarized and discussed.

IP17 Endocrine Disruptor Screening and Testing: Validation and Regulatory Implementation. Maciorowski, A. F.*, Timm, G. E., US EPA, Washington DC. The EDSTAC was convened in response to the Food Quality Protection Act and the Safe Drinking Water Act of 1996. To a large extent, EPA's implementation of endocrine disruptor screening and testing will follow the EDSTAC recommendations. However, a number of validation and regulatory implementation tasks will have to be accomplished prior to initiating endocrine disruptor screening and testing. A number of screens and tests require refinement, development, and validation prior to use in regulatory operations. Similarly, the Endocrine Disruptor Priority Setting Database msut be completed prior to soring and priority setting. Finally, there are legal and regulatory mechanisms that must be completed prior to actually requiring screening and testing. In this paper, we review EPA implementation steps and progress regarding the endocrine disruptor priority setting database development, the high throughput pre-screening (HTPS) demonstration project, the comprehensive HTPS project (15,000 chemicals produced over 10,000 pounds per year), and collaborative efforts to standardize and validate the endocrine disruptor screens and tests, and preparation of regulatory procedures for screening and testing.

IP18 Preliminary findings and Recommendations from the Ecological Committee on FIFRA Risk Assessment Methods: I. Process. Sunzenauer, I.M., US EPA, Washington, DC. In June 1997 the Office of Pesticide Programs began a new initiative to identify, develop, and validate tools and methodologies for predicting the magnitude and probabilities of adverse effects to nontarget species under the FIFRA regulatory framework. A key component of this initiative is the Aquatic and Terrestrial Workgroups or ECOFRAM, The Ecological Committee on FIFRA Risk Assessment Methods. These workgroups are comprised of experts drawn from government agencies, academia, contract laboratories, environmental advocacy groups, and industry and are identifying and developing probabilistic tools and methods for terrestrial and aquatic assessments. They are also identifying developmental information and validation needs to ensure that their approaches support an assessment process that is scientifically defensible. This interactive poster session will present ECOFRAM's preliminary findings and proposals.

IP19 Preliminary Findings of the Ecological Committee on FIFRA Risk Assessment Methods: II. Terrestrial Effects Assessment. R. S. Bennett, ecological planning and toxicology, inc., Corvallis, OR. The Terrestrial Workgroup of the Ecological Committee on FIFRA Risk Assessment Methods (ECOFRAM) was formed in June 1997 and met approximately monthly for a year developing tools and processes within the FIFRA framework for predicting the magnitude and probabilities of adverse effects to non-target terrestrial species resulting from the introduction pesticides into their environment. The Terrestrial Effects Subgroup, as an integral part of the Terrestrial Workgroup, identified and developed models to calculate dose-response relationships and their associated uncertainties, including the adequacy of current test methods for providing data and sources of intra- and interspecies variation. The Subgroup also evaluated methods for integrating exposure and effects data into probabilistic characterizations of risk and models for integrating risks to individuals into population level assessments. A report of the Subgroup's deliberations will be presented.

IP20 Preliminary Findings of the Ecological Committee on FIFRA Risk Assessment Methods: III. Terrestrial Exposure Assessment. E. Fite, US EPA , Office of Pesticide Programs, Washington D.C. The Ecological Committee on FIFRA Risk Assessment Methods (ECOFRAM) was formed in June 1997 and met approximately monthly for a year developing tools and processes within the FIFRA framework for predicting the magnitude and probabilities of adverse effects to non-target aquatic and terrestrial species resulting from the introduction of pesticides into their environment. The Terrestrial Exposure Subgroup, as an integral part of the overall terrestrial risk assessment, identified and developed models to estimate pesticide dose distribution in time and space to non-target terrestrial wildlife species taking in to account residue and animal behavior variables. The Terrestrial Subgroup also identified additional data required and developmental and validation steps needed to further improve non-target terrestrial pesticide exposure assessments under FIFRA. A report of the Work Group's deliberations will be presented.

IP21 Preliminary Findings of the Ecological Committee on FIFRA Risk Assessment Methods: IV. Terrestrial Risk Assessment. R. S. Bennett, ecological planning and toxicology, inc., Corvallis, OR and E. Fite, US EPA, Office of Pesticide Programs, Washington, DC. The Terrestrial Workgroup of the Ecological Committee on FIFRA Risk Assessment Methods (ECOFRAM) was formed in June 1997 and met approximately monthly for a year developing tools and processes within the FIFRA framework for predicting the magnitude and probabilities of adverse effects to non-target terrestrial species resulting from the introduction pesticides into their environment. The Terrestrial Workgroup identified and developed methods and models to quantify exposure and effects and their associated uncertainty. Risk characterization models were developed to estimate the magnitude and effects of pesticide use to terrestrial wildlife, along with quantification of uncertainties and identification of underlying assumptions. The Workgroup addressed the process for developing a probabilistic assessment under FIFRA, including the sequential steps and data required to support risk estimates for input into risk management decisions. A report of the Subgroup's deliberations will be presented.

IP22 Preliminary Findings of the Ecological Committee on FIFRA Risk Assessment Methods: V. Tools for Reducing Uncertainty in Aquatic Effects Analysis for Pesticides. Giddings, J.M.*, Springborn Laboratories, Wareham, MA, on behalf of the Ecological Committee on FIFRA Risk Assessment Methods (ECOFRAM). The Aquatic Effects Subcommittee of ECOFRAM has explored the use of a variety of tools and approaches for reducing uncertainty in the analysis of pesticide effects in aquatic risk assessments. (1) Generic life-table methods and age/stage models can be used to extrapolate from individual-based measurement endpoints such as survival, growth, and reproduction to population-based assessment endpoints such as recovery time and likelihood of local extinction; more complex, species-specific population models can be applied at higher tiers of a risk assessment. (2) Species sensitivity distributions can be analyzed to estimate risk to untested species and effects on community taxonomic richness. (3) Several approaches can be used to evaluate the effects of time-varying exposure (which is the typical case for many pesticides): time-to-event analysis of conventional toxicity data, extended laboratory toxicity tests using pulsed concentrations, uptake-depuration models coupled with dose-response models based on critical body burden, and population models to predict effects of repeated exposures. Other tools that can be used to address specific issues at higher tiers of a risk assessment include sediment toxicity tests, behavioral toxicity tests, and model ecosystems (microcosms and mesocosms). Much of the task remaining for the Subcommittee will be to integrate these tools into a logical sequential risk assessment process.

IP23 Preliminary Findings of the Ecological Committee on FIFRA Risk Assessment Methods: VI. Aquatic Exposure Assessment. P. Hendley, Zeneca Ag Products, Richmond, Ca. The Ecological Committee on FIFRA Risk Assessment Methods (ECOFRAM) was formed in June 1997. The Committee's purpose is to develop tools and processes within the FIFRA framework for predicting the magnitude and probabilities of adverse effects to nontarget aquatic and terrestrial species resulting from the introduction of pesticides into their environment. An Aquatic Exposure Subgroup was formed to identify probabilistic methods for aquatic exposure assessments and develop recommendations for future use by EPA. In addition, we are identifying information that must be developed in order to implement and validate the proposed methods to ensure that the proposed assessment process, if adopted by EPA, supports environmental decisions that are scientifically defensible. This poster describes the progress made by the Aquatic Exposure Subgroup. The proposed tiering system will be presented as the primary mechanism for making aquatic exposure assessments more predictable. Additional thoughts on a revised surface water exposure scenario selection process, a list of proposed changes in the Sub-Part N Environmental Fate Study Guidelines, lists of the key factors influencing aquatic exposure and details of the Tier IV landscape assessments will be given. Examples of a new PRZM-EXAMS post processor (RADAR) which analyses the temporal distribution of residue peaks will be provided. Finally, some information on the likely impact of the ECOFRAM process and draft recommendations for the future will be provided.

IP24 Preliminary Findings of the Ecological Committee on FIFRA Risk Assessment Methods: VII. Aquatic Risk Characterization and Tiered Risk Assessment Process. Giddings, J.*, Springborn Laboratories, Wareham, MA, and P. Hendley, Zeneca Ag Products, Richmond, CA. The Aquatic Exposure and Aquatic Effects Subcommittees of ECOFRAM have developed a tiered scheme for aquatic risk assessment of pesticides, consistent with the EPA's Framework for Risk Assessment, that integrates new developments in exposure and effects analysis. The lowest tier of the risk assessment process incorporates generic worst-case exposure modeling and a standard set of acute and chronic toxicity tests. Risk characterization at the lowest tier is based on risk quotients. At higher tiers, exposure is expressed probabilistically (see Poster V in this series), and effects are expressed in terms of population- and community-level assessment endpoints as well as individual-based measurement endpoints (see Poster VI in this series). Risk is characterized in the higher tiers as a function of probability of exposure and magnitude of ecological effect. Risk mitigation options (aimed at reducing exposure) are evaluated after each phase of the assessment. Results of risk characterization are conveyed to risk managers, who weigh the risks and benefits associated with each pesticide before making decisions on product registration.

IP25 Risk Based Corrective Action (RBCA) for Protection of Ecological Resources. Suedel, B.C., ENTRIX, Inc., Houston, TX; Biddinger, G.R., Exxon Company, U.S.A., Houston, TX; DeVaull, G.E., Shell Development Company, Houston, TX. ASTM Committee E50 on Environmental Assessment is developing a standardized approach to risk based corrective action (RBCA) for protection of ecological resources. The standard will outline an approach for managing chemical release sites by integrating site assessment, ecological risk assessment, risk management, and remedial action into a technically defensible framework. It will describe a logical sequence of activities and decisions to be followed from the time a release occurs until regulatory closure is achieved. The standard guide will incorporate a tiered approach, with increasing complexity at higher tiers. Each tier represents a level of effort that determines whether corrective action is appropriate and in which alternative action will eliminate or achieve an acceptable level of risk to ecological receptors. In early tiers, environmentally conservative assessments will be used to compensate for the relatively large uncertainty of using limited data and qualitative site information. In later tiers, site specific evaluations will be used to develop a more realistic exposure estimate, and refine the evaluation by focusing on ecological receptors and chemicals identified in previous tiers. The overall approach to developing the standard and the status of the standard at meeting time will be presented.

IP26 Comparative Analysis of Ecological Risk Assessment Guidance as Part of the ASTM Risk Based-Corrective Action Ecological Risk Assessment Standard. Costello-Walker, C.*, Langan Engineering and Environmental Services, Inc., Doylestown, PA; Menzie, C., Menzie-Cura Assoc., Chelmsford, MA; Kangas, M., Michael J. Kangas Risk Assessment/Management Services, University Heights, OH; Luke, N., Gradient Corporation, Somerset, NJ; Suedel, B., Entrix, Inc., Houston, TX; Swindoll, M., Exxon Biomedical Sciences, Inc., East Milstone, NJ; Meyer, A., Army Corp of Engineers, Montgomery, C., C.R. Montgomery and Associates, Arlington, MA; Hull, R., Beak Consultants, Ltd., Brampton, Ontario, Canada. More than twenty state and federal ecological risk assessment guidance documents were reviewed to support the development of an ASTM Standard for Risk Based Corrective Action (RBCA) for the Protection of Ecological Resources. This review had two primary objectives: 1) identify features to be included in the standard because they are commonly used and therefore represent important starting points and elements of consistency and, 2) identify limitations in existing guidance so that these critical issues would be included in the standard. Key issues identified by this review include: direction on how to identify *what is important to protect*, use of a tiered approach, role of benchmarks, role of exclusion criteria, what constitutes *significant ecological risk*, integration of ecological risk assessment with other aspects of risk management, impacts of scaling, how is site *recovery* evaluated under various levels of remedial response, risk communication tools, and how and when to involve stakeholders in the process. Also presented in this review is a summary of how the various guidance documents address specific issues related to either the development or implementation of the ecological risk assessment guidance. Finally, a complete listing of other pertinent Ecological Risk Assessment Guidance is presented in this review.

IP27 Screening Criteria in the RBCA Process for Ecological Receptors. Menzie*, C.A., Menzie-Cura & Associates, Chelmsford, MA; Roy, M. Radian Corporation, Austin, TX; Sorenson, M. Law Environmental, Kennesaw, GA. The ASTM Risk Based Corrective Action workgroup is developing a tiered assessment approach that is based in large part on the experience gained to date within state, provincial, and federal programs. One aspect of the guidance relates to the application of screening criteria to contaminated sites. Such criteria include a variety of methods that can be used at early stages of the risk assessment process to judge whether risks may be present and to take early action as needed. Benchmarks are among the most commonly used screening criteria and will be described in a separate paper in this series. In addition to the use of chemical benchmarks, criteria may include proximity of the site to valued habitats, measures of spatial and temporal scales of exposure, measures of *readily apparent harm*, various indicators of exposure, selected field observations, and selected measures of toxicity. The use of these approaches by various agencies will be discussed along with the strengths and limitations of these various methods. We will describe how and where these varied screening criteria can be incorporated into the RBCA process.

IP28 Risk Management Issues for Consideration When Applying RBCA to the Protection of Ecological Resources. Cheryl R. Montgomery*, C.R. Montgomery & Associates, Arlington, MA, E50.04.05 Task Group Members. Consistent with the provisional ASTM Risk-Based Corrective Action (RBCA) for Chemical Release Site, the ASTM *RBCA for the Protection of Ecological Resources* supports extensive interaction on the part of the responsible with concerned stakeholders at the very beginning of the process, and to continue this interaction through to site closure. In addition to historical and current technical data gathered in the course of a site assessment there are many risk management issues that should be considered throughout the process. It is these considerations which support the flexibility and broad applicability of the RBCA approach to site assessments. This poster outlines some of the issues which could be considered under three broad topics: 1) Technical Policy Decisions: What are they? What is their regulatory context? Why are they integral to problem formulation? 2) Site Management Dialog: What are the regulatory context(s) and the nature(s) of these discussions? What tools should be used to support problem formulation? and 3) Decision Points: What do they entail? Where and when in the RBCA process do they appear? What are the consequences of proposed remedial actions? How do these decision points interface with the human health risk assessment process? This poster will present a synopsis of the ASTM E50.04.05 task groups efforts to organize these considerations into the flexible tiered structure of the RBCA process.

IP29 Approaches to Scaling, Probabilistic Analysis and Handling Uncertainty in the Implementation of the ASTM Risk Based-Corrective Action Ecological Risk Assessment Standard. Kangas, M., Michael J. Kangas Risk Assessment/Management Services, University Heights, OH; Menzie, C., Menzie-Cura Assoc., Chelmsford, MA; Shibata, M. Tetra Tech, Lafayette, CA; Suedel, B., ENTRIX, Inc., Houston, TX. The contents of an appendix detailing the approaches to handling scaling issues, probabilistic approaches and uncertainty analyses are presented in support of the development of an ASTM Standard for Risk Based Corrective Action (RBCA) for the Protection of Ecological Resources. Approaches are provided for addressing spatial and temporal scaling of exposure factors and chemical, physical and biological sampling. Probabilistic approaches are presented to address handling site-specific sampling data, application of mathematical models (parameter and model error) and Monte Carlo Simulation. Approaches are provided for handling uncertainty in: problem formulation, development of data quality objectives, conceptual models and exposure scenarios. Recommendations and examples are provided on presentation of the results of uncertainty analyses. Separate approaches are provided for a conservative screening tier and site-specific assessments.

IP30 Selection and Use of Screening-Level Benchmarks for Risk-Based Corrective Action Ecological Risk Assessments. Swindoll, M.*, Exxon Biomedical Sciences, Inc., East Millstone, NJ; Hull, R.N., Beak International Inc., Brampton, ON., Saterbak, A., Shell Development Co., Houston, TX., Presiosi,D.V., Weinberg Group, Washington, DC., Sample, B., Oak Ridge National Laboratory, Oak Ridge, TN., Mazur, D. U.S. EPA, Region 5, Chicago, IL., Suedel, B., Entrix, Inc., Houston,TX. Guidance on ecological benchmark values is being prepared as an appendix to the ASTM Standard currently under development for Risk-Based Corrective Action for Ecological Risk Assessment (RBCA-ERA). Benchmarks are used as decision points for determining whether chemical concentrations in the environment have the potential to cause adverse impact to ecological resources. The RBCA-ERA benchmark appendix will provide the user with guidance on the selection and use of appropriate benchmark values for screening-level ERA activities. Specifically, topics to be addressed include: benchmark selection process; sources of existing benchmarks, with guidance for assessing the quality and appropriateness of benchmark values; benchmark limitations; and regulatory consideration. The appendix is intended to focus on selecting site-specific benchmarks rather than summarizing tables of published values. Also included are several user-friendly examples. This presentation will provide an overview of RBCA-ERA benchmark appendix.

IP31 Influence of Water Quality on Silver Toxicity to Rainbow Trout (*O. mykiss*), Fathead Minnows (*P. promelas*), and the waterflea (*D. Magna*): A Summary. Ownby, D.R.*, The College of William and Mary,Virginia Institute of Marine Science, Gloucester Point, VA, Karen, D.J., Clemson University, Pendleton, S.C, Cobb, G.P., Texas Tech University, Lubbock, TX, Klaine, S.J., Clemson University, Pendleton, S.C, La Point, T.W., Texas Tech University, Lubbock, TX. Silver is presently regulated in receiving waters using a hardness-based water quality criterion. The formula used to determine site specific limits is: Silver = $e^{(1.72[\ln(\mathrm{hardness})]-6.52)}$. This regression equation was derived using acute toxicity values for aquatic organisms and associated hardness levels. However, a hardness-based water quality criterion for silver in receiving waters may not adequately predict safe levels in natural waters. Data to adequately characterize silver toxicity to freshwater fish for combinations of a variety of water quality parameters have been lacking and are only presently being developed. We conducted toxicity bioassays to quantify water quality conditions under which silver, as silver nitrate, is toxic to *O. mykiss*, *P. promelas* and *D. magna*. Our 96 hour toxicity bioassays for *O. mykiss* indicate that chloride, hardness, and DOC protected against silver toxicosis, with DOC affording the highest protective effects. For *P. promelas* and *D. magna*, little protection was afforded by increased $CaCO_3$ alone, whereas DOC has a major ameliorating influence on measured silver toxicity. Lower concentrations of chloride (20 mg/L) had little effect on reducing silver toxicity. Results presented here indicate DOC is more important than hardness when predicting the toxicity of ionic silver in natural waters to *O. mykiss*, *P. promelas*, and *D. magna*. Mixtures of Cl^- and DOC show a synergistic protective effect. Thus, we suggest incorporating an organic carbon coefficient into the silver criterion equation.

IP32 Protective effects of dissolved organic matter against the physiological effects of waterborne silver on fish. Rose-Janes, N., and Playle, R.C*. Wilfrid Laurier University, Waterloo, Ontario, N2L 3C5. Full scale physiological experiments were run to investigate the protective effects of dissolved organic matter (DOM) against silver uptake by cannulated fish in soft water. Rainbow trout (*Oncorhynchus mykiss*, 300 g) were exposed to 0.1 $\mu mol \cdot L^{-1}$ Ag (as $AgNO_3$) in soft water in the presence and absence of added DOM. Partial protective effects of DOM against Ag entry into the fish and against the respiratory and ionoregulatory effects of Ag in soft water were seen with the addition of 8 mg $C \cdot L^{-1}$ dissolved organic carbon (DOC) isolated from Luther Marsh, Ontario. Full protective effects were seen with the addition of 35 mg $C \cdot L^{-1}$ DOC (Aldrich), so that a decrease in plasma Cl from 118 $mmol \cdot L^{-1}$ to 98 $mmol \cdot L^{-1}$ in the absence of DOM was eliminated. The amount of Ag accumulating on the gills over 90 h was reduced by the addition of DOM from 8.9 nmol $Ag \cdot g^{-1}$ wet gill tissue to 3.5 nmol $Ag \cdot g^{-1}$ wet gill tissue. The amount of Ag entering the plasma of the fish was also reduced by the Aldrich DOM, from about 1.6 $\mu mol \cdot L^{-1}$ Ag to 1.0 $\mu mol \cdot L^{-1}$ Ag.

IP33 Interactions between Magnesium and Silver in the Inhibition of Na^+/K^+-ATPase. Ferguson, Elizabeth A.*; Hogstrand, C., University of Kentucky, Lexington, KY. The Na^+/K^+-ATPase has been implicated as the key site of silver toxicity in physiological studies of Ag effects in freshwater fish. We have characterized the very potent inhibition of this enzyme by Ag *in vitro*. Results of Michaelis-Menten kinetics studies show non-competitive inhibition of the enzyme with respect to Na^+ and K^+ substrates and competitive inhibition with Mg^{2+} substrate. Such results diagnose that Ag must be interfering with the Mg-ATP or blocking the Mg^{2+} binding site on the Na^+/K^+-ATPase. Further analysis of the Ag and Na^+/K^+-ATPase interaction has shown that Ag has a very strong binding affinity for the Mg^{2+} binding site. The competitive inhibition of the enzyme with respect to Mg^{2+} has led to ongoing *in vivo* analysis of the significance of the Mg^{2+} concentration in the water for Ag toxicity. Na^+ uptake rates and Na^+/K^+-ATPase activity are being measured as an indicator of Ag toxicity at various Mg^{2+} concentrations. It is hypothesized that increased Mg^{2+} concentrations in the ambient water may act competitively with Ag for Na^+/K^+-ATPase binding and thus ameliorate Ag blockage of the enzyme thus decreasing toxicity.

IP34 Ag Toxicity to the Highly Tolerant European Eel (*Anguilla anguilla*) and the More Sensitive Rainbow Trout (*Oncorhynchus mykiss*). Grosell, M.*, Odense University, Denmark, Hogstrand, C., University of Kentucky, US, Wood, C.M., McMaster University, Ontario, Canada & Hansen, H.J.M., RISOE National Laboratory, Denmark. In soft water, 96h LC50 values were 160 and 20 µg Ag/l (as $AgNO_3$) for the European eel and rainbow trout, respectively. A nose to nose comparison of branchial ion transport (the key target for Ag toxicity) during acute exposure to Ag at 0.060 and 1.0 mM ambient Cl^- showed distinct species specific differences. 24 hours of exposure to between 10 and 100 µg Ag/l inhibited Na^+ influx by up to 95 % in a dose dependent manner in trout. Similarly Na^+ influx was inhibited by up to 80 % in eels after 30 hours of exposure. 1mM ambient Cl^- did not protect against the effect of Ag on Na^+ influx. Cl^- influx was inhibited by as much as 98 % in trout, also in a dose dependent manner. In contrast with Na^+ influx, Cl^- influx in trout was clearly protected from Ag toxicity by 1mM ambient Cl^-. The Cl^- influx in eels was only around 1 % or less of the corresponding influx in trout and was not affected by Ag. Ag exposure seems less severe to eels than to trout. Even though Na^+ influx was inhibited almost to the same extent in eels and trout, the much lower Na^+ uptake rate and thereby the lower whole body Na^+ turnover in eels means that the Na^+ pool is depleted at a lower rate during Ag exposure than in trout. Furthermore, eels seem to depend very little on branchial Cl^- uptake. This is in contrast to rainbow trout which have much higher branchial Cl^- uptake rates. (Supported by Kodak Canada Ltd and the NSERC IOR Program)

IP35 Acute and Chronic Effects of Silver Exposure on Egg Survival and Time to Hatch in Rainbow Trout *Oncorhynchus mykiss*. Brauner, C.J.* and Wood, C.M. McMaster University, Hamilton, Ontario. Early life stages in fish are often the most sensitive to various toxicants, however, only a few studies have examined the toxicity of silver in embryonic rainbow trout. The objectives of the present study were, firstly, to determine the effect of chronic exposure (starting just after fertilization) to 0.1 and 1.0 µgAg/l (added as $AgNO_3$) on time to hatch and % hatch, and secondly to screen for acute toxicity (48 h LC-50) of Ag during different developmental stages (every 4 days from fertilization) in rainbow trout eggs. Preliminary experiments indicate that at 14 °C in moderately hard water (120 mg/l, $CaCO_3$), chronic exposure to 0.1 µgAg/l in a flow-through system resulted in decreased time to reach 50% hatch (t_{50};23 days) but a slight increase total % hatch (87.6%) relative to control (t_{50};25 days, % hatch; 84.8%) and 1.0 ugAg^+/l (t_{50};25 days, % hatch; 79.5%) conditions. Ammonia, Na^+ and Cl^- content of the eggs were measured every 5 days during chronic exposure to silver. Preliminary studies indicate that eggs are most sensitive to acute Ag^+ exposure (as judged by a 48h LC50) 5 days following fertilization and then become increasingly more resistant up until hatch. (Supported by Kodak Canada Ltd and the NSERC IOR Program).

IP36 Silver in Marine Fish: Bioaccumulation and Physiological Response to Acute and Chronic Waterborne Exposures. Webb, N.A.* and Wood, C.M., McMaster University, Hamilton, ON, Canada. Various fish species were used to determine if silver bioaccumulation rates differed between marine teleosts (rainbow trout, plainfin midshipman, English sole, and tidepool sculpins) and elasmobranchs (spiny dogfish and long nose skate). The effects of salinity and exposure concentration on the bioaccumulation of silver in marine teleosts were also studied. Oxygen consumption, ammonia production and drinking rates were monitored to determine how internalized silver affects metabolic processes in marine teleosts. Our results show that the intestine plays a major role in the uptake of silver from the environment in marine teleosts (which drink the medium), while the gills are the only site of uptake in marine elasmobranchs (which do not drink the medium). Silver accumulation in teleost gills and intestines was 2-3 fold (over controls), while accumulation in elasmobranchs only occurred at the gills (5-6 fold over controls). The liver was the main site of silver bioaccumulation for both types of marine fish. Accumulation rates did not differ between species, but were dependent on both salinity and exposure concentration. Ammonia excretion was not affected during acute silver exposure, but both oxygen consumption and ammonia production were diminished during chronic exposure. Chronic silver exposure also affected drinking rates in marine teleosts. Drinking rates were decreased, with higher silver levels producing a greater effect. (Supported by Kodak Canada Ltd and the NSERC IOR Program).

IP37 Minimizing Ecological Impact from a Seafood Processing Facility. Rasnake, W.J.*; Gruber, D.; Biological Monitoring, Inc., Blacksburg, VA. Following the failure of several laboratory bioassays, this seafood processing facility was required to engage in a Toxicity Reduction Evaluation (TRE). The TRE implicated ammonia as the source for toxicity. Source reduction efforts lessened the ammonia levels and toxicity, although not sufficiently. Subsequent treatability studies indicated that nutrient deficiencies in the treatment plant were one cause for inadequate treatment. Although ammonia levels could be reduced by compensating for the nutrient deficiencies, toxic levels of nitrites would result. It was concluded that *Nitrobacter sp.* were either inhibited or not present in sufficient numbers to enable nitrite oxidation to nitrate. The present efforts at reducing toxicity are focused on bioaugmentation with commercially available bacteria and nutrient addition. Subsequent to treatment stability, laboratory bioassays and a field impact study will be engaged to assess the success of these efforts.

IP38 Aquatic Toxicity Associated with Drilling Fluids. Birkholz, D.A.*, B. Bayer, Enviro-Test Laboratories, Edmonton, AB., Canada, and G. Symonds, Amocco Canada Petroleum Company Ltd., Calgary, AB., Canada. Drilling fluid waste management and disposal in Alberta requires toxicity testing using luminescent bacteria, *Photobacterium phosphoreum*. A non-toxic response is required prior to surface disposal of the waste. Testing of a large amount of waste drilling fluid (4600 m^3) revealed a toxic response and as a result hindered disposal. Conventional treatment of the waste using potassium permanganate, activated carbon Nuchar S-A, and gypsum addition failed to render the waste non-toxic. Therefore, a toxicity identification evaluation (TIE) was initiated in order to isolate and identify the causative agent(s) for toxicity. This information was required to design and implement a detoxification plan for the waste in order to facilitate surface disposal. Application of the TIE revealed that the toxic agent(s) were effectively removed from the drilling fluid by solid phase extraction. Successive washes of the cartridge with water alcohol mixtures followed by toxicity testing with *Photobacterium phosphoreum* revealed that the toxic agent(s) were quantitatively recovered in two fractions containing 100% methanol. Both fractions were observed to be equally toxic. Analyses of these two fractions using gas chromatography/mass spectrometry and inductively coupled plasma mass spectrometry failed to identify the causative agents for toxicity. Additional analyses using liquid chromatography/mass spectrometry revealed the presence of polypropylene and polyethylene glycols. These compounds were found to be present in both fractions and readily explained the observed toxicity. These toxic compounds were readily degraded in the sample by the addition of nutrients, sewage seed and continous aeration for 5-days. Furthermore, this treatment successfully detoxified the sample. This procedure was successfully applied in the field and allowed for the surface disposal of the waste.

IP39 Toxicity Tracking: An Alternative to Exhaustive TIE Studies. Botts, J.*, Goodfellow, W., Routh, S., Spain, T., and Sohn, V. Aquatic Sciences Consulting, Woodbine, Maryland. Although TIE procedures can identify causes of nonpolar organic toxicity due to organophosphate pesticides, other nonpolar organic toxicants may be difficult to identify and confirm. POTWs in North Carolina often experience nonpolar toxicity that is related to chemicals used by textile industries. The components of this toxicity are complex and not well understood. TIEs may characterize dyes or surfactants as toxicants at these POTWs; however, identification and confirmation of the specific toxic compounds is limited by the lack of well-defined analytical techniques. If complex nonpolar organic toxicity is observed, municipal staff are advised to use an alternative approach developed by EPA that can rapidly and cost-effectively locate the sources of nonpolar organic toxicity. The Refractory Toxicity Assessment (RTA) procedure is described in the second edition of EPA's *Toxicity Reduction Evaluation Protocol for Municipal Wastewater Treatment Plants*, which is to be released later in 1998. RTA procedures have been applied in more than ten cases to identify sources of toxicity or assess the toxicity of planned discharges to POTWs. The approach involves treating sewer line or industrial user discharge samples in a simulation of the main POTW processes and testing the toxicity of the resulting effluents. A variety of POTW simulations have been applied, including conventional activated sludge, single-and two-stage nitrification, granular media filtration, and chemical addition. Inhibitory effects of the influent waste streams can be assessed simultaneously. RTA case examples include studies at the Cities of Reidsville and Henderson, NC. In 1992, RTA tests successfully identified an industrial user as a major source of chronic toxicity in the Reidsville collection system. Chemical optimization and waste minimization techniques implemented at the industry reduced effluent toxicity at the POTW from an average NOEC of 35% to ≥90%. Although the POTW effluent largely met the permit limit of an NOEC of ≥90% for nearly two years, effluent toxicity increased gradually from 1995 to 1997. Subsequent RTA studies are in progress to identify the source(s) of toxicity. RTA procedures also successfully identified sources of chronic toxicity at the City of Henderson POTW. Although control measures have been implemented, occasional effluent toxicity is still observed. The

City is currently using the chronic Microtox test to characterize additional sources of toxicity. Microtox results, which tend to correlate well for this effluent with results using the permit species (*Ceriodaphnia dubia*), will indicate areas of the collection system where additional RTA monitoring can be performed.

IP40 Toxicity Identification/Reduction Evaluation for the Canadian mining industry. Papineau, J.*, Natural Resources Canada, Ottawa, ON, Canada; L. J. Novak, R. L. Roy, K. H. Holtze, ESG International, Guelph, ON. The Aquatic Effects Technology Evaluation Program commissioned a study to evaluate and summarize the experience of Canadian mining industries with effluent Toxicity Identification Evaluations (TIEs) and Toxicity Reduction Evaluations (TREs). The objectives included an overall critical evaluation of the quality of TIE data, its benefits and limitations and, a survey to evaluate the utility of the TRE strategies addressing aquatic impacts from mining operations. Four case studies were selected based on two scenarios each with different outcomes: 1) the specific toxicant was identified and the toxic effluent was either eliminated or, not eliminated; 2) the general characteristics of the toxicant was identified and the toxic effluent was either eliminated or, not eliminated. Each reported study had varying degrees of *success/non success*. The outcome of case study #1 showed the Toxicity Identification (TI) /Reduction Evaluation (RE) process was useful to the copper/zinc mine; primary toxicants were identified but the presence of secondary toxicants was also suspected. The outcome of case study #2, a uranium mine, also demonstrated that the TI/RE process was useful; the primary toxicant was identified, modifications were made to the process (e.g. product substitution) and toxicity of the effluent was eliminated. The outcome of the gold mine in case study #3 determined the TI/TE process was not useful; the general characteristics of suspected toxicant(s) were identified, a full on-site treatment plan was built but the final effluent is still toxic. Case study #4, using a copper/nickel mine, showed the TI/RE process was not useful; the primary toxicant was identified but secondary toxicant(s) were not, a pH adjustment was added to the effluent treatment resulting in reduced toxicity.

IP41 Minimal Toxicity Necessary for an Effective TIE. Goodfellow, Jr., W.*, McCulloch, W., Sohn, V., and Hartzell, L. EA Engineering, Science and Technology, Inc., Sparks, Maryland When an effluent has been observed to be unacceptably toxic as part of the NPDES permitting process, a Toxicity Reduction Evaluation (TRE) is often required. A major tool for implementing a TRE is the Toxicity Identification Evaluation (TIE). Since toxicity of the effluent is necessary for performance of a TIE, the degree and consistency of the observed toxicity is a critical factor in the degree of success of the TIE. The authors have reviewed results from over 70 TREs, representing acute and chronic toxicity with freshwater and estuarine marine discharge situations over the last 10 years. The objective of this review was to develop information as to the minimal toxicity necessary for acute and chronic TIEs. As part of this paper, the results of TIE success in relationship to the magnitude of the toxicity is presented. This paper also presents recommendations and steps important to increasing the probability for a successful TIE.

IP42 Application of Antibody-Mediated Procedures to Identify Diazinon and Chlorpyrifos Toxicity and Bioavailability In Central California Surface Waters. Miller, J.L. and Miller, M.J., *AQUA*-Science, Davis, CA; de Vlaming, V., California State Water Quality Control Board, Sacramento, CA. Samples of surface water from the Alamo River and Orestemba Creek located in the California Central Valley were frequently acutely toxic to *Ceriodaphnia*. Phase I and II Toxicity Identification Evaluation (TIE) procedures, in conjunction with enzyme-linked immunosorbant assays (ELISAs), were conducted on the toxic samples. These studies identified two organophosphate (OP) insecticides, diazinon and chlorpyrifos, in many of the samples at concentrations which caused toxicity to *Ceriodaphnia* in laboratory water. An antibody-mediated chemical-specific procedure (F3), which selectively removes either diazinon (F3-D) or chlorpyrifos (F3-C) from aqueous samples, was applied to determine the relative role of the two OPs in the toxicity of the samples and to assess the bioavailability of the two OPs in the sample matrices. Using the F3 procedure, the toxicity of the sample was measured, the target OP was selectively removed using F3, and the sample toxicity was remeasured. The difference in toxicity before and after the F3 treatment was due to the OP that was selectively removed from the sample matrix. The F3 produced high (76-95%) removal of the target OPs with low (0-18%) removal of non-target pesticides. This highly selective removal permitted confirmation of the role of the two OPs in the toxicity of samples from both study sites. In addition, the F3 treatments showed that the bioavailability of both OPs varied widely in samples from Orestemba Creek and were 5-60% lower than in laboratory water. The F3 results showed that both OPs were consistently more toxic in the Alamo River matrix than in laboratory water.

IP43 Insecticide-Caused Toxicity in the Alamo River. de Vlaming, V.*, California State Water Resources Control Board, Sacramento, CA; DiGiorgio, C. and Deonovic, L., Aquatic Toxicology Laboratory, University of California, Davis, CA. Agriculture is the major land use in the Imperial Valley, CA. Almost 3 million acre-feet of Colorado River water are used to irrigate crops in this valley. The Alamo River, which is one of two major rivers in the valley, consists almost entirely of irrigation tailwater and discharges into the ecologically sensitive Salton Sea. Over 6 million pounds of pesticides are applied to crops in Imperial Co. every year. Water quality monitoring projects have been conducted on the Alamo River and Salton Sea from 1993 to the present. Water samples collected monthly at sites along the Alamo River were subjected to 96-hr. *Ceriodaphnia* toxicity tests and chemical analyses. Water samples elicited lethality on a seasonal basis--September through November and February/March. This pattern has been confirmed over several years of sampling. During these two periods essentially all samples from the entire length of the river, with the exception of a control site above the input of tailwater, caused 100% mortality. Chemical analyses of the toxic fall samples indicated that chlorpyrifos, diazinon, and malathion were responsible for the toxicity while diazinon, carbofuran, and malathion appeared responsible for the spring toxicity. Modified toxicity identification evaluations (TIE) were performed on 27 toxic samples over a 2 year period. TIEs demonstrated that toxicity was caused by metabolically activated organophosphorus insecticides in 25 samples. Data collected from 1996 to the present reveal that the Alamo River is discharging toxic concentrations of insecticides into the Salton Sea wildlife refuge.

IP44 Collaborative Effort to Develop Soil Screening Levels for Ecological Risk Assessment. Ells, S.J. and Wentsel, R., U.S. EPA, Washington, D.C., and Stahl, R., Dupont, Wilmington, DE. One of the first steps in the ecological risk assessment process involves screening of potential chemicals of concern, measured in the media of interest and compared to a predicted exposure level believed to pose minimal or no ecological risk. Those contaminants found at concentrations below the *acceptable* levels are eliminated from further evaluation; contaminants above these levels are evaluated further in the site-specific baseline ecological risk assessment. Currently, there are no peer-reviewed, ecologically based screening levels for contaminants in soil (eco-SSLs). Non-availability of eco-SSLs for soil poses considerable difficulty for EPA, other agencies involved in environmental regulation, and the regulated community. Consequently, EPA initiated an effort with the Chemical Manufacturers Association to form a workgroup involving representatives from Federal agencies, States, and the regulated community, whose goal is to develop soil screening levels for ecological endpoints. The resulting eco-SSL methodology could then also be used with site-specific data to develop risk-based protective soil cleanup goals. A steering committee and the following task groups have been formed within the overall Eco-SSL Workgroup to focus and coordinate research and data gathering efforts: toxic reference values, environmental chemistry, soil parameters, toxicity test method development, validation and field testing, risk characterization/integration, and exposure models. EPA plans to develop a consensus-based guidance, along with supporting models, equations, and rationale, that presents eco-SSLs for 25 chemicals and complex chemical mixtures often found in soil at hazardous waste sites at levels that could cause an ecological hazard:

DDT and metabolites, dieldrin, pentachlorophenol, TNT, total PAHs, total PCBs, TPHs, RDX, aluminum, iron, antimony, lead, arsenic, manganese, barium, nickel, beryllium, selenium, cadmium, silver, chromium, vanadium, cobalt, zinc, and copper.

IP45 Comparative Analysis of the Guiding Rationale and Modeling Concepts in the Derivation of Ecological Soil Screening Criteria in Europe, Canada, and the U.S. Rao, V*., Kulaga, K., Hartwell, J., Sukacz, A., Christian, T., Li, Y., Miller, D., DynCorp, Alexandria, VA; and Ells, S., U.S. EPA, OERR, Washington, D.C. EPA is in the process of developing soil screening levels (eco-SSLs) for ecological risk assessment. These screening criteria would serve as a tool in establishing preliminary baseline *unacceptable* levels of contaminants in soil for ecological endpoints. At present, only Washington State and Oak Ridge National Laboratories (ORNL) have developed eco-SSLs; a few others are involved in this process at varying levels. In the international scene, Canada and The Netherlands have published eco-SSLs. To define the guiding concepts and model parameters for a tool with nationwide application, a comparative analysis was performed on the six existing eco-SSL methodologies: (1) Washington State, (2) ORNL, (3) Canadian Council of Ministers of the Environment (CCME), (4) Ontario, (5) British Columbia, and (6) The Netherlands. Our analysis indicated that the existing methodologies placed varying emphasis on critical aspects of eco-SSLs. For instance, U.S. agencies in general have adopted a tiered approach in the derivation of eco-SSLs, in which toxicity parameters and elements of food-web pathways are combined for general or site-specific application. In contrast, Canadian agencies have attempted to address simultaneously both the protection of human and ecological receptors, and have based eco-SSLs on a consistent set of land-use patterns. Finally, the Dutch method has envisaged a unique quantitative method for applying information on soil chemistry and bioavailability of contaminants to a risk estimate, which is then combined with background data in the derivation of eco-SSL. However, none of these eco-SSLs adequately address ecotoxicity to higher members (mammalian and avian) of the ecosystem.

IP46 Soil Chemistry Issues in the Development of Ecological Soil Screening Levels. Wentsel, R. S.*, U.S. EPA, Washington, DC; Li, Y. R., DynCorp, Alexandria, VA, Menzie, C. A., Menzie-Cura & Assoc., Chelmsford, MA. Addressing complex soil chemistry issues is an important part of any process in the development of ecological soil screening levels for Superfund sites. This task links with other tasks under this effort to frame a process to develop soil screening levels for ecological receptors. It is hoped that the same process can be refined for application to site specific requirements. For the chemicals initially selected for evaluation, soil parameters (2-3 for each chemical) are identified that predominantly effect their availability/exchangeability. Initially a matrix using ranges of these parameters and qualitative (high, medium, low) ranges of availability/ exchangeability will be presented. Research efforts to refine and validate the matrix will be discussed. Further issues to address include: bioavailability, chemical concentration, time, toxicity to soil organisms, assessment endpoints, land use, relating to food web models, background levels of chemicals in soil, and nutrient requirements.

IP47 Ecotoxicity Test Methods Development for Ecological Soil Screening Levels. Checkai, R.T.*, U.S. Army Edgewood RD&E Center; Swindoll, M., Exxon Biomedical Sciences, Inc.; Kuperman, R.G., U.S. Army Edgewood RD&E Center; Stephenson, G. L., University of Guelph; Barclift, D., U.S. Naval Facilities Engineering Command - NorthDiv. Benchmark values for screening soils as part of Ecological Risk Assessments (ERAs) are needed to determine potential risk posed by contaminants of concern (COCs). An effort to develop peer-reviewed, ecologically-based soil screening levels (ESSLs) involves USEPA, DoD and other Federal Agencies, States, and industry representatives. A major task of the ESSLs initiative is to identify and develop chronic ecotoxicological bioassays; these bioassays will be used in developing soil test methodologies that are representative and sensitive indicators of potential site-specific, adverse ecological impact. The ecotoxicity tests will be used to develop ESSLs for use when toxic reference values (TRVs) are inappropriate or do not function adequately as protective endpoints. The toxicity tests may also be used to determine a protective endpoint by directly measuring site-specific contaminant effects. The status and results of this task group's efforts to develop standardized chronic toxicity tests for soils will be presented.

IP48 Wildlife Toxicity Reference Value Development for Ecological Soils Screening Levels. Hoff, D.J.*, U.S. EPA, Region 8, Superfund Technical Support, Denver, CO; Charters, D.W., U.S. EPA, Environmental Response Team, Edison, NJ; Rak, A., Air Force Center for Environmental Excellence, Consultant Operations Division, Brooks AFB, TX; Johnson, M., Army Center for Health Promotion and Preventative Medicine, Aberdeen Proving Ground, MD. A collaborative public and private effort is underway to develop methodologies needed to determine safe screening levels of contaminants in soils. One of seven tasks identified in this process is the development of wildlife toxicity reference values (TRVs) for an initial twenty five chemicals. Several steps are needed in this effort: 1) development of SOPs for searching toxicological data bases; 2) development of a searchable, maintained data base for wildlife toxicological literature; 3) development of SOPs for designating toxicological literature into qualitative levels of relative applicability for TRV extrapolation; 4) consensus on techniques for interspecific extrapolations of toxicologic endpoints. This is an ongoing project and this presentation is designed to solicit peer review from SETAC membership on the status and results in progress.

IP49 Standard Method for Evaluating the Quality of Toxicological Studies Used to Derive Wildlife Toxicity Reference Values. Brattin, W.J.*, ISSI, Inc, Denver CO, Hammon, T.L., ISSI, Inc, Denver CO, Hoff, D.J., U.S. EPA, Region 8, Superfund Technical Support, Denver, CO. A collaborative public and private effort is underway to develop Toxicity Reference Values (TRVs) for oral exposure of a variety of wildlife species to a number of chemicals commonly found in soil at Superfund Sites. One of the keys to success in this effort is the application of a standard procedure for evaluating and categorizing the quality and applicability of toxicity data presented in literature reports. This poster summarizes the proposed method which has been developed for application in this project. In brief, the evaluation procedure assigns a score to a number of key attributes of a study, including a) dose route (oral is ranked higher than inhalation or dermal), b) dose vehicle (intake in water or food is ranked higher than by gavage or capsule), c) dose range (studies that bracket the response threshold are ranked higher than those which do not establish both NOAEL and LOAEL values), d) study duration (chronic studies are scored higher than acute studies), e) measurement endpoints (growth and reproductive indices are ranked higher than severe effects such as lethality or subclinical effects such as biochemical or histological changes), and f) statistical significance (which in turn is a function of the number of animals and variability in measurement values within the study). The scores for the different aspects of the study are then combined into a single reliability index for that study, and this index is used in the process of deriving the TRV based on the weight of evidence across different studies.

IP50 Food chain and other models applied to the ecological risk of soil contaminants at Superfund sites. Stahl, R.G.*, DuPont, Wilmington, DE; Ells, S., USEPA Superfund Program, Washington, DC; Wentsel, R.S. USEPA, ORD, Washington, DC. In late 1997 a national work group was established to develop a process for setting ecologically-based, soil screening levels for the Superfund Program. A number of scientists from the public and private sector have joined this effort and divided themselves into seven task groups. Task group # 7 took on the challenge of assessing various effects and exposure models, including food chain analyses, for application in the soil screening process. The objective is to understand the models, their strengths and weaknesses, and when other approaches might be useful. The first meeting of the group resulted in the formulation of five basic questions that will drive the activities: 1) what models are available, 2) what are the *generic* inputs to these models (soil parameters, receptor type, exposure routes, etc., 3) what are the factors that change with site or which can be changed, 4) when are models useful and when are they not, and, 5) what other approaches might be used when models are not appropriate. In a preliminary analysis of a number of ecological risk assessments conducted for Superfund sites, we have found that a great diversity exists within the models that are used for food chain analysis. Some utilize one or two trophic levels, while others are highly complex and may extend up through four or five trophic levels. This finding along with preliminary results of what a *generic* food chain model may contain will be discussed.

PMP001 Investigation of the fate and distribution of alkylphenol surfactant metabolites in two Ontario sewage treatment plants. Bennie, D.T.* and Servos, M.R., Environment Canada, Burlington, ON; Schnell, A., Water Technology International Corporation, Burlington, ON. Over a two week period in 1997, comprehensive sampling and characterization studies were conducted at two STPs in Ontario. This involved a two-week and a two-day sampling campaign at plants A and B, respectively. As part of the characterization, samples of raw sewage, primary effluent, secondary/tertiary treated effluent, final disinfected effluent, raw combined sludge, anaerobically digested sludge, and secondary digester supernatant were collected and analyzed for various endocrine disrupting chemicals (EDCs). Among the EDCs of concern were the various alkylphenolics (e.g. alkylphenols, nonylphenol polyethoxylates and alkylphenolic carboxylates) and steroid hormones, such as 17ß-estradiol, ethenyl estradiol, and estrone. This study reports on a comparison of the reduction in alkylphenol loadings at the two facilities. Plant A is a state-of-the-art facility with conventional activated sludge treatment, including nitrification, followed by tertiary treatment and UV-disinfection. Plant B employs a conventional non-nitrifying activated sludge process and disinfection by chlorination. Utilizing the conservative conditions of Plant A, very low residual concentrations of total alkylphenols in final effluents (20-60 µg/L) were found despite the high influent concentrations of 500-3000 µg/L. Plant B exhibited good removals of alkylphenolics but lower efficiencies and somewhat higher residual concentrations, despite lower influent loadings than Plant A. Releases of alkylphenol polyethoxylate metabolites associated with the digested sludge were still significant and may cause concern regarding disposal of digested sludge.

PMP002 Methods for Determining Estradiol and Ethinylestradiol Concentrations in Water Samples. Snyder, S.A.*, Keith, T.L., Snyder, E.M., Kannan, K., Giesy, J.P.. Michigan State University, East Lansing, MI; Gross, T.S., USGS, Gainsville, FL. Several methods for identification and quantification of 17β-estradiol (E_2) and ethinylestradiol (EE_2) from water samples have been examined. In this study, water samples (5 L) were extracted *in situ* using Empore^{TM} solid-phase extraction (SPE) disks. The SPE disks were vacuum extracted and the extracts were concentrated to 1 ml. The extracts were fractionated to isolate the polar organics, including E_2 and EE_2 by normal phase high pressure liquid chromatography (HPLC). Reverse phase HPLC with fluorescence detection permits quantitation of E_2 and EE_2 to 25 ng/L of water. Radioimmunoassay (RIA) yielded quantitation of these compounds in the low pg/L range. E_2 and EE_2 were also derivatized using trifluoroacetic anhydride to polyfluorinated derivatives and analyzed by gas chromatography/mass spectrometry (GC/MS) and gas chromatography/electron capture detection (GC/ECD). Derivatization technique yielded detection limits in the upper pg/L range. It was determined that for the purpose of this study, RIA detection was most effective and sensitive. Several sites including sections of Lake Mead, Nevada, the Trenton Channel of the Detroit River, Michigan, and several municipal waste water treatment plants (WWTPs) in South-Central Michigan, influenced by waste water effluents were screened. Concentrations of E_2 ranged from nondetectable to several parts per trillion, while EE_2 concentrations ranged from nondetectable to nearly one part per trillion. The highest concentrations for E_2 and EE_2 were found in the effluent of a small WWTP in Southern Michigan with concentrations of 3.7 and 0.759 parts per trillion respectively.

PMP003 Characterizing pharmaceuticals in wastewater: A GC-FID screening method for estrogens. Tilton, F.*, Allgood, J.C., Schlenk, D., and Benson, W.H. Research Institute of Pharmaceutical Sciences (RIPS), The University of Mississippi, University, MS. The demonstration of cause and effect requires the integration of biological monitoring with chemical characterization. The production and use of Kg quantities of synthetic and natural estrogens used for contraception, hormone replacement therapy, and animal growth formulations in the U.S. may lead to the introduction of these pharmaceuticals to wastewater as both the parent and conjugated products. Specific assays such as radioimmunoassays are capable of quantifying only a few parent compounds in pg quantities with a high degree of specificity. Using Waters PrepPak 4000 radial compression - C_{18} Solid Phase Extraction (SPE) technology as a trace enrichment step, large volumes of water can be passed through columns to accumulate trace hydrophobic compounds of interest. Elution of the C_{18} columns with 50% methanol removes conjugates while subsequent elution with 100% methanol will remove parent compounds. Condensing samples to appropriate volumes using a roto-evap and filtering the methanol extracts (0.45um) prior to clean-up using silica gel/alumina columns removes contaminants within the retention time range of interest. Using a Hewlett Packard 6890 GC with a JW-1701 (30m x 250um) column and a Flame Ionization Detector (GC-FID) allows for detection of various estrogenically active compounds in the ng/L range with >90% recovery. Estradiol, its two major metabolites estriol and estrone; equillin used in hormone replacement therapy, ethinylestradiol, diethylstilbestrol, testosterone, as well as their conjugates elute with a MDL in the low ppb range depending on the specific compound. This method is utilized along with GC/MS verification to screen wastewater for estrogenically active compounds of varying biological activity in order to identify the total estrogen content.

PMP004 Endocrine Disruption in Largemouth Bass in a Re-flooded Agricultural Area. Douglas, D. R., Johnson, W. E., Florida Game and Fresh Water Fish Commission, Eustis, FL; Denslow, N., The University of Florida, Gainesville, FL; Wieser, C., Wiebe, J., and Gross, T. S., U. S. Geological Survey, Gainesville, FL. Long Farm (300 ha), one of five re-flooded farms adjacent to Lake Griffin in Central Florida, was utilized for muck-farming from the early 1960s through 1992 when it was flooded for restoration. Long Farm was stocked in April 1993 with 81,500 largemouth bass. Of the five former farms, Long Farm had the highest pesticide concentrations in soil and fillet samples, and poor bass reproductive success. This study was undertaken to evaluate seasonal and age-related trends in reproductive hormone levels in individual bass (tagged) and to determine if successful reproduction would occur if adult fish were transported to hatchery ponds. Blood plasma samples were analyzed for estrogen (E), 11-keto testosterone (11-KT) and vitellogenin. The E/11-KT ratio was found to be reversed (11-KT > E) in 81% of the females samples in 1995 (N = 32), 53% in 1996 (N = 51), and 15% in 1997 (N = 27). Reversed ratios in males were found in 11% of the 1995 samples (N = 36), 50% in 1996 (N = 60), and 17% in 1997 (N = 63). Reversed E/11-KT ratios were found in 1% of the male and 3% of the female bass in Lake Woodruff, a reference lake. Five female bass were each paired with two male bass in individual hatchery ponds in March 1997. All females had a history of reversed E/11-KT ratios, but spawned within the first week, and approximately 5,000 to 12,000 fry were collected from each pond. Largemouth bass collected and aged from Long Farm in 1998, indicated limited spawning or survival in 1995 and 1996. A need exists to further define the relationship among pesticide contamination, habitat and water quality, and fish reproduction in agricultural restoration areas.

PMP005 Examination of Reproductive Function in White Sucker Populations on the Moose River Basin Influenced by Hydroelectric Dams and Pulp and Paper Effluent. McMaster, M.E.*, Jardine, J.J. and Munkittrick, K.R. Environment Canada, Burlington, Ontario, Canada; Portt, C. C. Portt and Associates, Guelph, Ontario, Canada; Oakes, K. and Van Der Kraak, G.J., University of Guelph, Guelph, Ontario, Canada. Starting in 1991, we began to examine white sucker (*Catostomus commersoni*) populations within the Moose River drainage basin in Northeastern Ontario, initially focussing on two pulp mills. These studies identified alterations in gonadal development and corresponding reductions in plasma sex steroid hormone levels. More in-depth studies have continued, confirming our earlier findings of reproductive alterations in fish downstream of the mills. Studies have followed reproductive function as the pulp mills have modernized their pulping and waste treatment processes. This system however, has also been altered by the addition of a number of hydroelectric dams. Our more recent studies are examining reproductive function in white sucker populations upstream and downstream of both run-of the-river dams in which water flow is not controlled, and peaking facilities that hold large amounts of water for times in which demand for electricity are high. Preliminary examination of the data indicates alterations in gonadal development in fish collected downstream of the peaking facility. A complete assessment of reproductive function will be made in fish collected from this river system.

PMP006 Estrogenic Effects of Pesticides Used to Control Mosquitos in Galveston, County, TX. McDaniel, W.C. and Howard, C.L. University of Houston-Clear Lake, Houston, TX. Currently, there is a great deal of research focused on the possible reproductive effects that many types of pollutants, including pesticides, may have on wildlife. The goal of my research is to examine the possible estrogenic effects of pesticides used by the Galveston County Mosquito Control District. Wild-caught, male Gulf killifish (*Fundulus grandis*) will be exposed to the organophosphate malathion and the synthetic pyrethroid resmethrin under salinity regimes using induction of vitellogenesis as the test end-point. Vitellogenin will be separated by polyacrylamide gel electrophoresis and quantified using an imaging densitometer. The assay will be validated by radioimmunoassay or enzyme-linked immunosorbant assay (ELISA). If either assay shows estrogenic activity, experiments will be conducted to determine the time course of vitellogenesis and the maximal response as measured by peak levels of circulating vitellogenin.

PMP007 Estrogenic Potential of Organic Contaminants in New York-New Jersey Harbor Complex Sediments. *McArdle, M.B.*, McElroy, A.B., and Biskus, A.A., State University of New York, Stony Brook, NY. Laboratory studies have shown that many of the hydrophobic organic contaminants (HOCs) found in urban sediments such as polynuclear aromatic hydrocarbons, polychlorinated biphenyls, pesticides, dioxins, and alkylphenols are known to act as endocrine disruptors and exert various reproductive effects on aquatic organisms. To determine whether NY-NJ Harbor sediments contain estrogenic substances, a laboratory study is being performed. Vitellogenin production will be measured in naive male *Fundulus heteroclitus* injected with organic extracts of NY-NJ Harbor sediments by Western blot analysis. A field study is also being conducted to determine whether or not there is evidence of endocrine disruption in fish collected from the NY-NJ Harbor Complex. Vitellogenin production will also be assessed in males of resident *(F. heteroclitus)* and in juveniles of migratory *(Morone saxatilis* and *Pomatomus saltatrix)* fish. Results of the laboratory study will indicate whether NY-NJ sediments are a potential source of endocrine modulating compounds. The field study will furnish data on whether migratory and/or resident fish species in NY-NJ Harbor show signs of hormonal alterations. Based on the results obtained in these studies, further studies may be necessary to provide valuable data on the NY-NJ Harbor to managers of this area.

PMP008 Endrocrine Disrupting Potential of Various Flavonoid Compounds. Kiparissis, Y.* and Metcalfe, C.D., Trent University, Peterborough, ON, Canada.; Niimi, A.J., Fisheries and Oceans, Burlington, ON. The endocrine and reproductive processes of fish exposed to pulp and mill effluents are impaired. The responsible chemicals could be either chlorinated by-products or natural plant constituents released during pulping operations. Preliminary analyses using LC-MS indicated the presence of flavonoid-type compounds in wood pulp and pulp mill effluents. The assessment of various flavonoids as endocrine modulators was determined with the *in vitro* recombinant yeast assay and with *in vivo* Japanese medaka (*Oryzias latipes*) models. Flavone, chrysin, apigenin (flavones), flavonol, galangin, kaempferol, quercetin (flavonols), flavanone, naringenin (flavanones) and (+) catechin (flavan-3-ol) induced the development of testis-ova, an intersex condition, in medaka exposed to nominal concentrations ranging from 10 ppb to 1000 ppb for 3 to 5 months. In addition, the expression of the secondary sex characteristics was either reduced or altered in 20 to 40% of medaka exposed to the same flavonoids. Embryotoxicity data indicated that the unsubstituted flavonoid ,flavone, flavonol and flavonol, as well as the pentahydroxy-substituted quercetin were the most toxic at ppb nominal concentrations. The yeast assay indicated that flavone, apigenin, galangin and kaempferol have the ability to weakly bind to the estrogen receptor. These data support the argument that phytochemicals such as flavonoids have endocrine disrupting potential and may be partly responsible for the adverse reproductive effects observed in feral fish downstream of pulp mills.

PMP009 Development of a Screening Assay for Endocrine-Disrupting Chemicals using the Fathead Minnow. Kahl, M.D.*,Jensen, K.M.,Korte, J.J.,Pasha, M.S., Linnum, A.L.,Ankley, G.T., U.S. EPA, Duluth, MN. The fathead minnow, *Pimephales promelas*, has been identified as a suitable test organism for use in screening endocrine-disrupting chemicals (EDC). Test methods, focused upon endpoints such as lethality, growth and reproduction, for this species have been well documented. However, endpoints proposed for the EDC screening assay are relatively more specialized, focusing, for example, on vitellogenin induction, changes in concentrations of plasma steroid hormones (testosterone, 11-ketotestosterone, β-estradiol), gonadal histology, secondary sexual characteristics and behavior. To incorporate these types of endpoints into standard assays with the fathead minnow requires research focused on appropriate measurement techniques, as well as careful characterization of baseline conditions. In this presentation, we describe our studies in these areas. We also will present studies with fathead minnows characterizing the use of subcutaneous injections with corn oil, cocoa butter, as carriers for ECD whose water solubility or stability precludes exposure via the water.

PMP010 Effects of Methyl Mercury on Plasma Estrogen and 11-Keto Testosterone in Nile Tilapia (*Oreochromis niloticus*). *B.S. Arnold**, C.H. Jagoe, and R. Reinert, University of Georgia, Athens Georgia and T.S. Gross, USGS BRD, Gainesville, FLA. Studies have focused on factors influencing Hg bioaccumulation by fish but few have examined effects of realistic Hg burdens on fish themselves. To better understand effects of low levels of methyl mercury in fish, Tilapia, *Oreochromis niloticus*, were dosed with CH_3Hg using intraperitoneally implanted capsules designed to dissolve over 180 days. Pellets contained either 0.1 (low dose) or 1.0 (high dose) mg of CH_3HgCl_2, placebos contained only the carrier matrix. Blood samples taken prior to dosing and for seven months after were split for mercury and hormone analysis. After one month high dose fish had a mean blood mercury level of 0.48 ppm (dry wt) and low dose fish had 0.1 ppm. By the second month mercury levels decreased to 0.29 ppm in the high dose group and 0.06 ppm in the low dose fish. Initially, females in all treatments had estrogen/11 keto testosterone (E/T) ratios of 3.2 to 3.6, and males 0.5. E/T ratios in high dose females declined to 0.7 after one month of exposure, then increased slightly to 1.4. In the same treatment, male E/T ratios increased to 1.0 in the first month and 2.3 by the second month. Similar responses occurred in the low dose females where E/T ratios declined to 2.0, and in low dose males where ratios increased to 1.1 by the second month of treatment. E/T ratios in the treated fish differed significantly from pretreatment means, and from control fish. These results indicate that methyl mercury affects plasma hormone concentrations in tilapia, and that mercury can act as an endocrine disruptor and potentially impair reproduction in fish.

PMP011 Effluent Exposure and Alterations in Steroid Hormone Metabolic Elimination from the Fathead Minnow (*Pimephales promelas*). Parks, Louise G.* and LeBlanc, Gerald A., North Carolina State University, Raleigh, NC. Steroid hormone metabolic clearance pathways are susceptible to changes (i.e. induction or suppression) resulting from xenobiotic exposure. Altered rates of hormone clearance may impact steroid hormone homeostasis and steroid hormone-dependent processes such as development and reproduction. In the present study, the metabolic elimination of testosterone was characterized in 95 day old fathead minnows (*Pimephales promelas*) and the susceptibility of these processes to change resulting from exposure to an industrial effluent was evaluated. Fathead minnows eliminated a variety of testosterone metabolites including oxido-reduced, hydroxylated, and conjugated derivatives. Prolonged exposure of fathead minnows to an industrial effluent significantly increased the rate of elimination of testosterone as androstenedione and as two testosterone metabolites tentatively identified by GC/MS as androstanetriols. Analyses are underway to establish whether the enhanced metabolic elimination of testosterone following effluent exposure resulted in a decrease in serum testosterone or an increase in serum androstenedione levels.

PMP012 Sex steroid kinetics in rainbow trout, *Oncorhynchus mykiss*, following benzo[a]pyrene administration. Kennedy, C.J.* and Smyth, K., Department of Biological Sciences, Simon Fraser University, Burnaby, BC. Xenobiotic exposure has been suggested as one cause of reproductive failure in fishes leading to the elimination of populations in a contaminated area. These effects may occur by several mechanisms depending on the contaminant, which can include alterations in sex steroid hormone concentrations in circulation. It is still unclear if a link exists between the induction of biotransformation enzymes by prior contaminant exposure, altered sex steroid levels and reproductive failure in fish. In this study, rainbow trout were administered 50 mg/kg of benzo[a]pyrene via intraperitoneal injection and the levels of circulating testosterone and estradiol monitored daily for one week. Benzo[a]pyrene injection increased the activity of ethoxyresorufin O-deethylase activity up to 7 fold, while cytochrome P450 levels remained unchanged. The levels of circulating testosterone and estradiol were decreased in both mature male and female trout for 6 days up to 50% and 62%, respectively. In order to begin to determine the mechanism of plasma steroid concentration depression in benzo[a]pyrene-exposed fish, trout were administered an intraarterial dose of 3H-estradiol via a cannula implanted in the dorsal aorta. The blood concentration-time course of estradiol was fit to a two-compartment open toxicokinetic model. No significant differences were seen between control and benzo[a]pyrene-injected fish in any of the determined toxicokinetic parameters (e.g. half-life of the injected steroid). These results continue to illuminate the relationship between PAH and mixed function oxidase inducers on sex steroid kinetics, and the mechanism of altered plasma steroid kinetics in fish exposed to these compounds.

PMP013 An Evaluation of Atrazine as a Potential Endocrine Disruptor in Largemouth Bass. Grady, J., University of Florida, Gainesville, FL; Wieser, C.*, Wiebe, J. and Gross, T.S., USGS-BRD, Florida Caribbean Science Center, Gainesville, FL. Previous reports from this laboratory have indicated endocrine-disrupting effects for the water soluble herbicides atrazine and 2,4-D in largemouth bass. However, these initial studies did not utilize eco-relevant doses nor exposures. The current study was designed to evaluate potential endocrine disrupting effects of atrazine in largemouth bass at eco-relevant doses and exposures. Adult, reproductively active, largemouth bass were randomly distributed into 4 treatments: 0, 25, 50 and 100 ug/l for 20 days (15 male and 15 female per 400 gal treatment/tank). Tanks were checked daily and water quality monitored bi-weekly. Fish were removed from each tank, at the completion of exposure, weighed, measured, blood and gonadal tissues collected. The following parameters were used to evaluate the effects of atrazine on reproduction: sex steroid/hormone concentrations, vitellogenin, gonadal somatic indices (GSI), and gonad histology. In general, health status was not impacted by atrazine treatment. For male bass, plasma sex steroid concentrations were not altered by the 25 or 50 ug atrazine/l treatments, whereas the high treatment (100 ug/l) increased plasma 11-ketotestosterone concentrations. For female bass, the low treatment of 25 ug atrazine/l did not alter plasma sex steroid concentrations, whereas both the 50 and 100 ug/l treatments increased plasma estradiol concentrations. These data suggest endocrine responses of largemouth bass to eco-relevant atrazine doses and exposures.

PMP014 An Evaluation of Methyl Mercury as an Endocrine Disruptor in Largemouth Bass. Fynn-Aikins, K., USFWS, Vero Beach, FL; Gallagher, E., University of Florida, Gainesville, FL; Ruessler, S., Wiebe, J., Gross, T.S. USGS BRD, Gainesville, FL. Previous studies have primarily focused on the fate and transport of mercury in aquatic species, however, sublethal effects have not been routinely documented. Mercury has been routinely listed as a potential endocrine disruptor in wildlife with little or no supporting data. Adult largemouth bass (n= 240) were randomly distributed into four treatments: 0, 1.56, 3.12 and 6.25 mg methyl mercury chloride per kilogram of diet (30 male and 30 female per treatment). Prior to dosing, 10 fish (5 male and 5 female) were collected from each treatment tank for the collection of blood and tissue samples to determine baseline levels of mercury. Additional fish (n=10, 5 male and 5 female) were collected from each treatment biweekly. Blood samples were utilized for the analysis of sex steroid hormones (estradiol and 11-keto-testosterone) and vitellogenin. Gonadal tissues were collected for histological evaluation of sex and reproductive status. Liver tissues were collected for glutathione transferase and mRNA analyses. Tissues were collected from each fish for mercury analyses. Plasma estradiol and 11-ketotestosterone concentrations were not altered by the low dose (1.56 mg methyl mercury chloride/kg diet) treatment regardless of sex. However, plasma 11-ketotestosterone concentrations were decreased in male largemouth bass exposed to the 3.12 and 6.25 mg methyl mercury diets. Similarly, plasma estradiol concentrations in female bass were decreased following exposure to the 3.12 and 6.25 mg methyl mercury diets. Certain GST catalytic activities were also inhibited by high doses methyl mercury exposure. These results indicate that dietary exposure to methyl mercury affects plasma hormone concentrations in largemouth bass, and that mercury can act as a endocrine disruptor and potentially impair reproduction in fish.

PMP015 Documentation of Endocrine-disruption and Altered Seasonal Reproductive Parameters for Brown-bullhead Catfish from Several Lakes in Central Florida. Denslow, N.D.*, Kroll, K., Schoeb, T. University of Florida Gainesville, FL; Johnson, B., Florida Game and Freshwater Fish Commission, Eustis, FL; Wiebe, J., Wieser, C., Gross, T.S., USGS-BRD, Gainesville, FL. Previous efforts from this laboratory have documented altered endocrine function and sexual differentiation for largemouth bass from reclaimed agricultural sites in Central Florida. These reclaimed lake sites have been exposed to a variety of contaminants which are potentially endocrine-disrupting. A year long, seasonal, survey of brown-bullhead catfish populations was conducted on Lakes Apopka, Griffin, and Woodruff as well as from two reclaimed muck-farm sites: Knight Farm and Long Farm to examine reproductive function and potential relationships to environmental contaminants. Plasma samples were collected from approximately 20 fish (10 males and 10 females) monthly from each lake for one full calender year. Plasma samples were analyzed for estrogen, testosterone, 11-ketotestosterone and vitellogenin concentrations. Additionally, fillet samples were collected from 5 female brown-bullhead catfish from each site and analyzed for pesticide levels. Fish from Lakes Apopka, Griffin and Woodruff had similar seasonal patterns. In contrast, catfish from the reclaimed muck-farms, Knight and Long Farm ponds (a highly contaminated site) had greatly reduced estrogen and testosterone concentrations as compared to all other sites. Seasonal cycles were minimal in catfish from Knight Farm and absent for fish from Long Farm. A survey of age classes for brown bullhead catfish indicated probable reproductive success on both Farm pond sites. These studies indicate altered reproductive endocrine function for brown-bullhead catfish living in contaminated lakes and demonstrate the necessity for associated characterization of seasonal reproductive cycles and reproductive endpoint success.

PMP016 Endocrine Disruption in Largemouth Bass and Environmental Contaminants: A Reconnaissance Study of the Lower St. Johns River Basin. Gross, T.S. *, Wieser, C., Kernaghan, N., Gross, D.A., USGS-BRD, Gainesville, FL; Sepúlveda, M. S., University of Florida, Gainesville, FL; Johnson, W.E., FL Game and Freshwater Fish Com, Eustis, FL; Higman, J., St. Johns River Watyer Management Diatrict; and Holm, S., Georgia Pacific Corp., Atlanta, GA. Previous results from our laboratories have indicated altered endocrine parameters for largemouth bass at the St. John's River - Rice Creek confluence. These alterations may be related to local exposure to chemical components within papermill effluents. Therefore, a multi-site study was conducted on Rice Creek and several main-stream and tributary sites throughout the lower St. Johns River basin. Seven collection sites within the St. Johns River mainstream with differing contaminant profiles were utilized for this survey. Three collection sites within the Rice Creek drainage basin were also utilized. Each of the ten sites were surveyed during March 1998 (reproductive season). Largemouth bass (10 male and 10 female) were collected from each site. Fish were weighed, measured, blood collected, and each animal sacrificed for the conduction of a full health assessment. The following parameters were used to evaluate reproductive effects: sex steroid/hormone concentrations, vitellogenin, gonadal somatic indices (GSI), gonad histology, and size/number of eggs in females. General health was assessed using several blood parameters and complete necropsies. Organ somatic indices, organ histology, and liver EROD activity were also determined. Reproductive and health assessments indicated significant differences between sites. Significant alterations in endocrine parameters were observed at several sites. Plasma sex steroids, gonadal weights and GSI were depressed for largemouth within Rice Creek as compared to other streams in the same drainage basin. These results indicate endocrine related anomalies in largemouth bass exposed to papermill effluents and other environmental contaminants within the lower St. Johns River basin.

PMP017 Lake Apopka Revisited: An Evaluation of Environmental Contaminants and Reproductive Anomalies in Alligators. Giroux, D., The University of Florida, Gainesville, FL; Percival, H.F., Rice, K., Wiebe*, J., and Gross, T.S., USGS-BRD, Gainesville, FL. Environmental contaminants have been hypothesized as a primary contributing factor in the decline of juvenile alligators on Lake Apopka, as well as the associated reproductive anomalies. Alligator eggs from Lake Apopka have greatly reduced hatchabilities and increased neonatal mortalities. The current study was designed to examine DDT and its metabolites (i.e. DDE) as potential causes of reproductive and neonatal anomalies. Adult female, nesting, alligators from lakes Apopka (n=29) and Woodruff (n=10) were trapped for egg, fat biopsy and plasma collection. Fat samples were utilized for contaminant analyses. Egg clutches were monitored for: total eggs, nested eggs, hatched eggs, viability rate, hatch rate, neonatal mortality, survival rate and production rate. Concentrations of DDE were higher for maternal fat and eggs from Lake Apopka alligators. Total egg numbers were increased for Lake Apopka clutches, however, the number of eggs nested were decreased as were the hatched eggs, neonatal survivors, nest rate, viability rate, hatch rate, production rate and survival rate. In spite of these differences, there was not any significant correlation between clutch characteristics and DDT nor DDE concentrations in maternal fat or eggs. However, when clutches and females from Lake Apopka were separated into two geographical populations, North and South, significant differences were observed: clutches from South Lake Apopka had increased nested eggs, hatched eggs, neonatal survivors, viability rate, hatch rate and production rate than clutches from North Lake Apopka. These results indicate two distinct populations and effects for Lake Apopka alligators. Results do not support the earlier hypothesis which linked DDT or DDE to clutch anomalies on Lake Apopka. Additional studies are needed to examine other contaminants.

PMP018 Use of rainbow trout (*Oncorhynchus mykiss*) and brown trout (*Salmo trutta*) to screen Hamilton Harbour water for estrogenic effects. Gamble, A.V.[1]*, Bennie, D.[2], Brown, S.[2], Hodson, P.V.[3], Karrow, N.A.[4], Parrott, J.[2], Servos, M[2], Solomon, K.R.[1], Sherry, J.P.[2] [1]University of Guelph, Guelph, ON, [2]Environment Canada, Burlington, ON, [3]Queen's University, Kingston, ON, [4]University of Waterloo, Waterloo, ON. Hamilton Harbour is highly impacted by both industrial and domestic waste. It receives sewage treatment plant (STP) effluent from several communities including the cities of Burlington and Hamilton. There has been much recent concern over the potential estrogenicity of some components of STP effluents including natural estrogens and the alkylphenols and their degradation products. We assessed the ability of Hamilton Harbour waters to induce vitellogenin (Vg) production in juvenile fish. Vg was measured by ELISA. In the first of three studies, rainbow trout were caged at six sites in Hamilton Harbour and L. Ontario. Vitellogenin was induced in fish caged at sites in the Windermere Arm of the harbour. Those sites are within the zone of influence of Hamilton STP effluent and of several large industrial sites. In a second study, one group of rainbow trout were caged in the effluent mixing zone of the Burlington STP and another group were concurrently exposed to a dilution series of effluent from the Burlington STP in the laboratory. Vitellogenin was induced in the laboratory exposed fish by 50% effluent, but not in the field exposed fish. In the third experiment, rainbow and brown trout were caged in the Windermere Arm of Hamilton Harbour and in L. Ontario.

PMP019 The effect of three androgens, DHEA, androstenedione, and dihydrotestosterone, on vitellogenin levels in rainbow trout, *Oncorhynchus mykiss*. Shilling, A.D.*, Williams, D.E., Toxicology Program and Marine/ Freshwater Biomedical Sciences Center, Oregon State University, Corvallis, OR. The contribution of androgens to vitellogenin (Vg) production in rainbow trout, *Oncorhynchus mykiss*, was investigated. Androgens may affect Vg levels by: (1) directly binding the estrogen receptor thereby inducing or competitively inhibiting Vg gene transcription; (2) indirectly altering estrogen metabolism; or (3) acting as precursors for estrogens, which induce vitellogenin production. In this study, juvenile trout were fed either dehydroepiandrosterone (DHEA), 4-androstene-3,17-dione (androstenedione) or a non-aromatizable androgen, 17β-hydroxy-5α-androstane-3-one (dihydrotestosterone) at doses ranging from 0.05 - 50 mg/kg/day for two weeks. Blood plasma Vg and 17β-estradiol levels were determined using a specific enzyme-linked immunosorbant assay (ELISA) and an enzyme immunoassay (EIA), respectively. Comparisons were made intrasex only, because sex differences for Vg and 17β-estradiol were significant in controls for all three treatment groups ($p < 0.05$). In DHEA treated males, significant 5-fold increases in Vg and 17β-estradiol were observed at the 50 mg/kg dose ($p < 0.05$). Conversely, dihydrotestosterone did not induce Vg significantly in males and significantly decreased 17β-estradiol by 55% ($p < 0.05$, t-test) at 50 mg/kg and in females, actually decreased Vg by over 50 % at doses above 0.5 mg/kg ($p < 0.05$). This suggests an indirect androgenic mechanism, possibly altering hormonal feedback loops, resulting in the down regulation of estrogens. Significant increases of total cytochrome P450 content in liver microsomes by DHEA above 0.5 mg/kg and at 5 mg/kg androstenedione support a hypothesis that androgens may affect steroid metabolism. Perhaps androgens influence Vg levels by playing a role in estrogen homeostasis acting as precursors for estrogens or increasing estrogen degradation via P450 upregulation. These studies demonstrate the need to explore androgenic mechanisms and their contribution to estrogenic and antiestrogenic responses observed in rainbow trout. (Supported by NIH grants ES-07060, ES 03850 and ES 04766).

PMP020 Development of a vitellogenin ELISA that is broadly cross-reactive among fish, amphibians and reptiles. Foret, P., Selcer, K.W. Duquesne Univ., Pittsburgh, PA and Palmer, B.D.*, Univ. of Kentucky, Lexington, KY. Vitellogenin has proven to be a sensitive biomarker for the exposure of lower vertebrates to environmental endocrine disruptors. There is a growing interest in the ability to quantify vitellogenin in a diversity of species using antibody-based techniques, such as ELISAs. However, vitellogenins show significant variation in amino acid sequence among species. Therefore, antibodies produced using vitellogenin from a single species are often poorly cross-reactive with vitellogenins from other species. Although antibodies could be developed for each species, it would be advantageous to have an antibody that cross-reacts against vitellogenins from a number of species. Our laboratory has developed a monoclonal antibody that has cross-reactivity across a broad range of vertebrate vitellogenins. The peptide sequences of vertebrate vitellogenins (representing organisms from four classes of vertebrates: Agnatha, Osteichtheyes, Amphibia, and Aves) were retrieved and aligned in order to identify conserved regions. A sequence of 23 amino acids was selected due to its highly conserved nature and antigenic properties. A synthetic peptide was produced and used as antigen in Balb/C mice. Hybridomas were produced, positive clones expanded and ascites fluid produced using standard procedures. The resulting monoclonal antibodies have been tested against a diversity of vertebrate species for their utility in quantifying vitellogenin. To date, these antibodies prove cross-reactive with vitellogenins from various species, including fish (teleosts), amphibians (anurans) and reptiles (chelonians and squamates). These data provide a strong indication that our monoclonal antibodies are broadly cross-reactive with vertebrate vitellogenins. These antibodies should prove useful not only in the detection and screening of environmental endocrine disruptors, but in comparative endocrinology and reproductive studies as well. Supported by NIH ES07621 and USDA 9504243.

PMP021 Effects of Xenobiotic Chemicals on Vitellogenin Induction in the Sheepshead Minnow, *Cyprinodon variegatus*. Hemmer, M.J.*, Folmar, L.C., Webb, B.L., U.S. EPA, Gulf Breeze, FL; Kroll, K., Denslow, N., University of Florida, Gainesville, FL. Temporal and dose-response relationships of plasma vitellogenin (Vtg) induction were established for sheepshead minnows (*Cyprinodon variegatus*) treated with the putative xenoestrogens p-nonylphenol (p-NP), methoxychlor and endosulfan. Thirty-two adult male fish per treatment were continuously exposed to aqueous concentrations of 1, 10, 20, 40 and 80 µg p-NP/L, 1.5, 3, 6, 12 and 24 µg methoxychlor/L, and 25, 50, 100 and 200 ng endosulfan/L using an intermittent flow-through dosing apparatus. Separate triethylene glycol (50 µl/L) and estradiol (100 ng/L) treatments served as the negative and positive controls, respectively. Four fish were randomly sampled from each concentration on days 2, 5, 13, 21, 25, 42 and 49 of exposure and serum Vtg levels determined by direct ELISA. P-NP induced a dose dependent increase in plasma Vtg over the entire time course of exposure with significantly elevated Vtg levels by day five at nominal concentrations ≥10 µg p-NP/L. Exposure to 1.0 µg p-NP/L resulted in highly variable tonic plasma Vtg levels of <6 mg /ml. The highest exposure concentration of methoxychlor resulted in 75% mortality within six days of exposure. Dose-dependent Vtg induction occurred in the remaining concentrations with the 3 and 6 µg methoxychlor/L treatments exhibiting maximal Vtg levels at 21 days of exposure, then decreasing to slightly lower tonic levels for the remainder of the 49 day test. Only the 12 µg methoxychlor/L treatment demonstrated a linear increase in plasma Vtg over time. Exposures with endosulfan failed to induce measurable plasma Vtg at the concentrations tested.

PMP022 Vitellogenin in Wild Male Flounder, *Pleuronectes yokohamae*, **in Tokyo Bay.** Hashimoto, S.*, Tokyo University of Fisheries, Tokyo, Japan; Bessho, H., Tokyo University of Fisheries, Tokyo, Japan; Sato, K., Tokyo University of Fisheries, Tokyo, Japan; Hara, A., Hokkaido University, Hokkaido, Japan; Iguchi, T., Yokohama City University, Kanagawa, Japan; Nakamura, M., Teikyo University, Tokyo, Japan; Fujita, K., Tokyo University of Fisheries, Tokyo, Japan; Otsuki, A., Tokyo University of Fisheries, Tokyo, Japan. Concerns about environmental estrogen are widespread, because of their potential to cause deleterious physiological effects in humans and wildlife. Estrogenic chemicals including nonylphenol, polychlorinated biphenyls, and DDE are found in sewage treatment effluent, river water, and estuaries in Tokyo Bay. Fishes may take up such contaminants. The objective of this study was to determine the extent of any observed effect of environmental estrogen on marine fish by examining the occurrence of vitellogenin (VTG) in male serum. The sandwich fluorometric EIA was developed for analysis of flounder VTG with antiserum against lipovitellin (Lv), along with VTG isolated from wild flounder (Pleuronectes yokohamae). The lowest measured concentration was about 2 ng/ml. Flounders were collected from Tokyo Bay and off the coast of Hokkaido (control) from March 1997 to March 1998. The results indicate that VTG occurs in male flounder in Tokyo Bay throughout the year. High concentration of VTG was detected (maximum concentration: 670 ng/ml) in 30 of 71 wild male flounders in Tokyo Bay. The presence of VTG in male flounder throughout the year suggests that estrogenic effects on wild fish are occurring over a wide portion of Tokyo Bay and are not limited to areas affected by sewage effluent.

PMP023 Assessment of estrogenic activity in rockfish and halibut exposed to crude oil. Hwang, I.Y.* and Lee, E.K., Dept. of Environemental Sci., Inje University, Korea. Massive oilspill caused to ruine marine ecosystem for a long time after Sea Prince Oilspill in Korea, 1995. As a part of screening processes to assess sublethal effect of crude and its weathered oil on marine biota, induction of blood Vg in fish was measured after treatment with either test oil alone, test oil plus 17β-Estradiol(E2), or E2 alone in the laboratory. Each 10 male black rockfish (*Sebastes schlegeli* Hilgendorg) and halibut (*Paralichthys olivaceus*) were repeatedly injected 3 times as 3 day interval with oil at 200mg/kg body weight. At 8th day, E2 (2mg/kg) or carrier (150ul of alcohol:tween-80=1:1 v/v) was treated to fish. And then, the amount of Vg in whole blood was analyzed by SDS-PAGE/densitometry and compared to the sham control at 10th day. Crude oil slighly elivated the amount of Vg in both kinds of fishes, but its weathered oil did not. The level of Vg induction in rockfish was similar to that in halibut. However, there was no difference of Vg level in blood between oil plus E2 and E2 alone treated groups. In this study, we found the SDS-PAGE/densitometry may be useful to determine the level of Vg in whole blood.

PMP024 Rounding Up the Facts about Rodeo: Evaluating Estrogenic Effects of Associated Surfactants. K. M. Kubena*, B. Smith*, M. Tagal, C. E. Grue, Washington Cooperative Fish and Wildlife Research Unit, University of Washington, School of Fisheries, Seattle, WA. Efforts to control Spartina in Washington State have been hampered by concerns over the potential non-target effects of Rodeo, the only herbicide approved for use on Spartina in Washington, and the potential effects of the surfactants used with Rodeo. The active ingredients of two commonly used surfactants, R11 and X77, are alkylphenol ethoxylate compounds previously shown to have estrogenic effects on rainbow trout, Oncorhynchus mykiss. This study evaluated the potential estrogenic effects of three surfactant formulations used in combination with Rodeo on rainbow trout. Growth (length and weight), and vitellogenin induction were evaluated. Surfactant concentrations included 300 and 3000 µg/L (ppb). No statistically significant effect on growth was seen in fish treated with the Rodeo tank mixes, but we feel this effect could be masked by the fact that the fish varied greatly in size. Vitellogenin induction was not observed in fish treated with the Rodeo tank mixes. Efforts are currently underway to repeat the experiment using the higher concentration with fish of a more standardized size.

PMP025 The Effect of 4-Nonylphenol on *Gambusia affinis. Vinturella, A.E.*, Hopkins N. E., Mullin, A.H., Cheek, A.O., Alworth, W.L., and Bart, H.L., Jr., Tulane University, New Orleans, LA. Nonyphenol is a byproduct of a common industrial surfactant; it is a known estrogen mimic in vertebrates, causing the production of vitellogenin (an egg yolk precursor) in male oviparous teleosts. For this study, groups of mosquitofish, *Gambusia affinis*, were exposed to two sediment concentrations (25 ppm and 250 ppm) of 4-nonylphenol for 40 days. Over the 40 day period, concentrations of 4-nonylphenol were monitored in sediments and water. Homogenized fish tissue was electrophoresed on an SDS-PAGE (6%) and assayed against fathead minnow vitellogenin antibody to determine the induction of vitellogenin by 4-nonylphenol. Water concentrations of 4-nonylphenol were minimal and did not differ significantly between treatments. Sediment concentrations remained constant over the treatment period. Two major bands of estrogen-induced proteins (approximately 120 and 31 kDa) were found to comigrate with two fathead minnow vitellogenin bands. In the males, there was a significant, dose- related difference in the densities of these bands. A third band was found at 73 kDa. None of the mosquitofish bands crossreacted with the fathead minnow vitellogenin antibody. Western blots will be repeated with other vitellogenin antibodies as they become available. Due to the uniformly low concentration of dissolved 4-nonylphenol in the water samples, it is proposed that the route of uptake of 4-nonylphenol by fish is through adsorption to fine suspended sediment particles that enter the fish's system. Once in the system, 4-nonylphenol was shown to exhibit a dose- and sex-dependent inductive effect on the production of two proteins that are similar in molecular weight to vitellogenin, a biomarker for estrogenic compounds.

PMP026 Effects of endocrine disrupters in the fathead minnow (*Pimephales promelas*). *G.H. Panter*[1], P.M. Campbell[2], T.H. Hutchinson[3], D.E. Kime[4], R. Länge[5], P. McCahon[6], J.P. Sumpter[7], L.J. Tattersfield[8] and C.R. Tyler[7]. [1]EMSG c/o Brixham Environmental Laboratory, Zeneca Limited, Freshwater Quarry, Brixham, Devon, United Kingdom, TQ5 8BA; [2]Procter & Gamble, Belgium; [3]ZENECA Limited, UK; [4]Sheffield University, UK; [5]Schering AG, Germany; [6]Rhône-Poulenc Cedex Agro, France; [7]Brunel University, UK; [8]Shell Research & Technology Centre, UK. The Endocrine Modulator Steering Group (EMSG) of the European Chemical Industry has developed an aquatic research programme, with input and support from several academic and industry research groups. The first EMSG research phase is aimed at developing a short-term *in vivo* test for detecting endocrine disrupters in juvenile fish (protocol based on the OECD Test Guideline 204). The fathead minnow was selected as the test species in view of the existence of a validated ELISA for vitellogenin (VTG) analysis and the species' proven use in other areas of endocrine disrupter research (eg evaluation of sewage effluent oestrogenicity and steroidal activity). Juvenile fish (approx. 150 mg wet weight) were exposed by continuous immersion to individual test chemicals for up to 21 days at 25°C. The test chemicals included natural and synthetic substances, incorporating (anti-) oestrogenic and androgenic activities and also aromatase inhibitors. After 4, 7, 14 and 21 days exposure, fish were subsampled and evaluated for biochemical effects which might be indicative of endocrine disruption. The primary endpoint was whole-body VTG levels, while additionally, male and female sex hormones (oestradiol, 11-ketotestosterone and testosterone) were also determined in whole-body homogenates. The presentation will summarise the available results on a range of reference chemicals in support of a short-term *in vivo* fish test for detecting endocrine disrupters.

PMP027 Early Life Stage Effects of PCBs or Mercury on *Fundulus heteroclitus*. Matta, M.B., NOAA, Seattle, WA; C. Cairncross, NOAA, Seattle, WA; J. Linse, EVS Environment Consultants, Seattle, WA; P. Peronard, U.S. EPA, Atlanta, GA; L. Francendese, U.S. EPA, Atlanta, GA; R. Kocan, University of Washington, Seattle, WA. A laboratory study was conducted to determine tissue concentrations in adult fish that are associated with reductions in survival and fecundity in exposed adults, fertilization success and hatch success of eggs, and early life stage survival of offspring. Fish were collected from Sapelo Island, GA, and brought to Seattle, WA where they were fed commercially available fish food contaminated with either methyl mercuric chloride or Aroclor 1268 dissolved in acetone. Treatment groups included fish treated with four concentrations of each contaminant, uncontaminated controls, and solvent controls. Exposure concentrations in whole bodies included 0.05-12 µg/g wet weight for mercury and 0.01-15 µg/g for Aroclor 1268. At least ten breeding pairs were established in each treatment group. Male fish treated with mercury suffered significant mortality. Although the study was not designed to investigate behavioral effects, males treated with mercury appeared to be much more aggressive than control males or those treated with Aroclor 1268. Mortality in mercury treated males appeared to be related to this aggression, with more submissive fish being attacked and killed by dominant fish. This effect was observed in male fish with a mean whole body total mercury concentration of 0.47 µg/g and greater. Female fish exposed to the highest concentration of mercury (where the mean whole body concentration was 12 µg/g wet weight) had reduced fecundity when compared to other treatment groups. No statistically significant effects were observed in fish treated with Aroclor 1268.

PMP028 Effects of Mercury on Reproductive Success of Fathead Minnows. Hammerschmidt, C.R.*, University of Wisconsin-La Crosse, La Crosse, WI; Sandheinrich, M.B., University of Wisconsin-La Crosse, La Crosse, WI; Wiener, J.G., U.S. Geological Survey, La Crosse, WI; Rada, R.G., University of Wisconsin-La Crosse, La Crosse, WI. Relatively little research has addressed the effects of mercury on populations of fish. Methylmercury causes gonadal abnormalities and impairs gonadal growth and development in male teleosts; however, the major effect on reproductive success may result from toxicity of maternally derived mercury to embryos and larvae. Fish embryos in the wild are exposed to methylmercury primarily via maternal transfer during oogenesis. We examined (1) the effects of maternal and dietary burdens of mercury on clutch size, hatching success, concentrations of mercury in the eggs, and on growth and survival of larvae; and (2) the effects of paternal and dietary burdens of mercury on rate of fertilization and hatching success. We manipulated mercury concentrations in four groups of fathead minnows (*Pimephales promelas*) by feeding each group a diet contaminated with 0, 2, 4, or 10 µg methylmercury · g^{-1} dry weight until they reached sexual maturity. During gametogenesis, we separated mating pairs from each group into subgroups and manipulated the mercury content of diet of each subgroup. We measured concentrations of mercury in the parental fish and offspring, and clutch size, hatching success, growth, and survival of larvae. Preliminary results indicate that larval growth and mercury concentrations in the eggs and adult fish differed among groups. This study will provide insight on the effects of mercury on reproduction in fish.

PMP029 Interference with Oogenesis in Xenopus laevis by Cadmium. Lienesch, L.A.*, Dumont, J.N., Bantle, J.A., Oklahoma State University, Stillwater, OK. Cadmium (Cd) is reproductively and developmentally toxic. It accumulates in the ovaries of Xenopus and thus potentially affects not only oogenesis but also future embryonic development. This study focused on determining the effects of Cd on Xenopus laevis oogenesis and the subsequent development of embryos. Cadmium chloride ($CdCl_2$) at 0.5, 0.75, 1.0, 3.0 or 5.0 mg/kg body weight, was injected into the dorsal lymph sacs of sexually mature female Xenopus every other day for 21 days. Control frogs were injected with 0.7% saline. At the end of the exposure period, females were divided into 2 groups. One group of females was killed, their ovaries removed, weighed, and a portion of the ovary analyzed for Cd by Inductively Coupled Plasma - Mass Spectrometry (ICP-MS). Oocytes were also removed from an ovarian lobe using collagenase, staged according to oocyte diameter, and scored for morphologic characteristics. The other group of females was bred to untreated males and the Frog Embryo Teratogenesis Assay-Xenopus (FETAX) was run. Results of $CdCl_2$ exposure indicated that ova from females exposed to 3.0 mg or 5.0 mg Cd/kg were morphologically different from the control group. Ova from females exposed at 0.5 mg/kg showed no apparent morphological changes. Cadmium significantly reduced the percentage of stage II - IV ova at all exposures (P = < 0.02). Results from the ICP-MS analysis indicated that the ovary significantly accumulated $CdCl_2$. Preliminary FETAX results indicated growth inhibition in progeny from exposed females. Further experiments will analyze the amount of Cd in fertilized eggs and embryos and further examine transgenerational effects. By combining the data from reproductive toxicity testing and FETAX, susceptible periods in the Xenopus life cycle will be identified and the effects of reproductive disruptors on oogenesis and development determined.

PMP030 The Role of Vitellogenin in Maternal Transfer. Monteverdi, G.H.*, Duke University, Durham, NC, DeVito, M.J., U.S. EPA, RTP, NC, and R.T. Di Giulio, Duke University, Durham, NC. The presence of vitellogenin (VTG) significantly alters the distribution and quantity of lipophilic compounds in fish serum. Analyses of serum from estradiol (E2)-treated versus untreated male *Fundulus heteroclitus (Fundulus)* exposed by i.p. injection to [^{3}H]-2,3,7,8-trichlorodibenzo-p-dioxin ([^{3}H]-TCDD) or [^{14}C]-benzo(a)pyrene ([^{14}C]-BaP) showed marked difference in both serum protein and radiolabel profiles. In the absence of VTG, radiodetctor chromatograms present a single peak, associated with a large protein peak of $T_{elution}$ = 50min (Peak 3). Chromatograms of serum from E2-induced males show two radioactivity peaks: one associated with VTG (Peak 2), and one associated with the major protein peak (Peak 3) observed in uninduced males. Similar results were observed in experiments with [^{14}C]-BaP. The identity of the Peak 3 is unknown, though it is likely another serum lipoprotein based on its propensity for binding both TCDD and BaP. Correspondence between vitellogenesis and tissue distribution characteristics of [^{14}C]-BaP and [^{3}H]-TCDD were also examined. In the BaP studies, it was found that [^{14}C]-BaP injected i.p. into gravid *Fundulus* is rapidly sequestered in growing oocytes; detectable quantities appearing in ovary samples within 4 hours. The greatest quantities of BaP were observed in the gall bladder, while detectable levels remained in blood, liver and filet samples throughout the 15-day (360 hour) timecourse. DES-treated fish showed greater concentrations of BaP in blood (all time points) and ovary (5 of 6 time points) samples, suggesting a correlation between circulating VTG levels, and rates of serum binding and sequestration of BaP. Similar results were obtained in studies with [^{3}H]-TCDD. Ongoing tissue distribution studies include the examination of the relationship between oocyte maturational state (size) and uptake of the radioligands.

PMP031 Development, validation and application of a highly responsive reporter gene assay for estrogenic and anti-estrogenic compounds. Legler, J., Agricultural University Wageningen, NL; Van der Burg, B., Hubrecht Laboratory, Utrecht, NL; Vethaak A.D., National Institute for Coastal and Marine Water Management/RIKZ, Middelburg, NL; Brouwer, A., Murk, A.J.* Agricultural University Wageningen, NL. Development of an estrogen-receptor mediated chemical activated luciferase reporter gene expression (ER-CALUX) assay was attempted by stable transfection of luciferase reporter genes in a number of cell lines. Stable transfection of 3xERE-Luc construct in the T47D breast cancer cell line resulted in an extremely sensitive, highly responsive cell line. Following 24 hour exposure to estradiol, stably transfected T47D.Luc cells demonstrated a detection limit of 0.5 pM, EC50 of 6 pM and a maximum response at 30 pM of 100-fold relative to solvent controls. No clear reduction in responsiveness has been found over extended culture (50 passages). Anti-estrogens ICI 182,780 and tamoxifen inhibited luciferase induction when tested with co-administration of estradiol. Diethystilbesterol, genistein, o,p'DDT and nonylphenol were the most potent (pseudo-)estrogens tested. Determination of interactive effects of (pseudo-)estrogens showed that, in combination with 3 pM E2, pseudo-estrogens were additive. Slightly more than additive effects were found for combinations of methoxychlor and chlordane, o,p'DDT and nonylphenol, and dieldrin and endosulfan. In testing environmental mixtures of chemicals, polar acetone extracts from sediments demonstrated higher estrogenicity than nonpolar hexane extracts. High estrogenic activity was found in effluent extracts from wastewater treatment plants, but also surface water from some Dutch rivers contained measurable estrogenic activity. The extract from 5-50 ml water was enough to determine estrogenic potency with the ER-CALUX method.

PMP032 Development of a rainbow trout recombinant chimeric receptor reporter bioassay to screen for the potential estrogenicity of environmental contaminants in fish. Clemons, J.H.*, Matthews, J.B. and Zacharewski, T.R. Michigan State University, East Lansing, MI. Recently, the U.S. Environmental Protection Agency has supported the use of estrogenic screening assays for both mammalian and wildlife species in order to elucidate the contribution of environmental contaminants in the endocrine disrupter issue. Our lab has established an *in vitro* rainbow trout (*Oncorhynchus mykiss*) bioassay to screen for compounds that act through the rainbow trout estrogen receptor. The assay utilizes a recombinant chimeric receptor consisting of the Gal4-yeast DNA-binding domain linked upstream to the ligand-binding domain (A.A. 214-576) of the rainbow trout estrogen receptor (Gal4-RtDEF). This cDNA is co-transfected with a Gal4-regulated luciferase reporter gene that is expressed in the presence of ligand-bound Gal4-RtDEF. The transfection efficiency and performance of the Gal4RtDEF and Gal4 reporter gene was evaluated in 3 fish cell lines, PLHC-1, RTG-2 and RTH-149 and the human breast cancer cell line, MCF7. A comparison of the ability of Gal4RtDEF and the human chimeric Gal4-estrogen receptor (Gal4HEG0) to bind 17β-estradiol (E2) in the MCF7 cells revealed that the human receptor had approximately an order of magnitude greater sensitivity for E2 than the chimeric fish receptor. This was also reflected when two different fish cell lines, PLHC-1 and RTG-2, were transfected with either the fish or the human chimera. The PLHC-1 appeared to be the best cell line for transfection efficiency and inducibility; however, all three cell lines appeared to respond similarly when different estrogenic xenobiotics were added to the bioassay. This *in vitro* fish recombinant receptor bioassay is an excellent tool for fast, accurate, cost-effective screening of environmental contaminants or mixtures for estrogenic activities in fish.

PMP033 The development of a harmonised approach to the ecological risk assessment of plant protection products. Laskowski,D ,Dow Agrosciences, Indianapolis,IN, USA; Arnold,D.J*, AgrEvo UK Ltd. Saffron Walden, Essex, UK. This paper has been prepared by the European and American Crop Protection Associations in response to the growing need to seek a harmonised approach to ecological risk assessment.This is of particular concern in the context of the registration and use of plant protection products (PPP's) where regulatory decision making is outpacing the development of tools to do the job and where probabilistic exposure and effects asssessments are likely to replace current predominantly deterministic approaches. It is recognised that it is neither possible ,nor desirable to have one globally harmonised ecological risk assessment for plant protection products as their varied use patterns guide the levels and extent of exposure and hence their consequential effects . Rather,the approach to ecological risk assessment presented in this paper is in the form of a framework that may be used as a model for the construction of specific risk assessments. From a global perspective this has one important advantage. The framework can account for the different levels of resources and data available in a particular situation because it does not prescribe what is needed as building blocks at specific points. It therefore allows the user to apply the framework in a way that meets local circumstances.The scheme describes the stepwise nature of assessment through issue identification,risk characterisation and risk refinement . Whilst the scheme focusses on the types of data/information that may be appropriate for different levels of the ecological risk assessment it also requires that the consideration of benefits , and a risk /benefit analysis are an inherent part of the process. With OECD interest in developing a harmonised approach to the risk assessment of chemicals it is hoped that the ECPA/ACPA scheme will provide them with an appropriate framework .

PMP034 Evaluation and Elucidation of Coproduct presence and persistence in technical malathion used during the 1997 Florida Medfly Eradication Project. Sherblom, P.M., University of South Florida, Tampa, FL. Between early June and late August 1997, Central Florida suffered an infestation by the Mediterranean fruit fly (medfly). The eradication program relied on localized ground treatment and aerial spraying of a malathion laced bait. At one point the aerial spray program encompassed 425 square miles of urban, suburban and agricultural areas in three counties. These applications caused concern about the public health impact of not only the malathion, but also of the coproducts which can be present in technical grade malathion. Though the coproducts are less than three percent of the malathion mixture, some have greater health effects. Citizen groups raised specific concerns about the potential presence of isomalathion, malaoxon and diethylfumerate. Citizens were also concerned about the relative persistence of the coproducts and their potential formation during weathering of the malathion bait. Two investigations were undertaken to evaluate these issues. In the first, coproduct presence and abundance were evaluated in six *weekly* subsamples of the technical grade malathion used in the spray program. Hexane dilutions of the neat product were analyzed using gas chromatography with flame ionization or mass spectral detection. The second investigation evaluated the relative persistence of the coproducts to that of malathion. In this work, neat product was stored at 25 and 35 degrees Celsius for up to three months prior to determination of the coproduct abundance. In addition to trace quantities of isomalathion and malaoxon, two additional unexpected compounds were present, tentatively identified as diethylmalonate and a tri-sulfur malathion analog. During storage only isomalathion showed a definate increase relative to malathion. This increase occured at both storage temperatures, but was greater at the higher temperature.

PMP035 Treatment of Pesticide-Contaminated Raw Water: Importance to Drinking Water Exposure Assessment. Pisigan, Jr., R.A., U.S. EPA, Washington, DC. A study was conducted to gather information on the removal and transformation of some pesticides in the water treatment systems and afterwards evaluate how the treatment operations could potentially affect water exposure assessment. Eleven pesticides (lindane, dieldrin, chlordane, heptachlor, alachlor, pentachlorophenol or PCP, parathion, fenitrothion, fenthion, demeton-S-methyl, atrazine) were used in the assessment. Contaminated raw water have been subjected to lime coagulation, carbon adsorption, aeration, and disinfection with chlorine (Cl_2) and ozone (O_3). The removal efficiencies were found to vary with each pesticide and pesticide class, and other factors related to dosage, contact time, and chemical composition of the raw water. High lime treatment removed about 13 to 70% of dieldrin, chlordane,and organophospahtes. The removal efficiency of packed tower aeration were low (0 - 29%) for lindane, PCP, and chlordane. Higher removal efficiency (70 - ~100%) were obtained using granular activated carbon (GAC) and biologically activated carbon (BAC) for the chlorinated pesticides. Chemical disinfection generally gave satisfactory removal (70 - ~100%) for some pesticides. Chemical treatment with ozone did not only remove pesticides but also transform them into products with different and possibly unknown toxicity compared to that of the parent compound. Aldrin and heptachlor have been demonstrated to be converted by ozone to dieldrin and heptachlor oxide, respectively. Parathion was transformed in the presence of ozone to products that include paraoxon, 2,4-dinitrophenol, and picric acid. Seven ozone byproducts were identified in natural water with atrazine. Some of the major products were deethylatrazine, deisopropylatrazine, and 2-chloro-4,6-diamino-s-triazine. Transformation products generated from either ozone or chlorination, not the pesticides in raw water, are important in conducting human health risk assessment of drinking water.

PMP036 Estimation of Pesticide Residues on Weed Seeds for Wildlife Risk Assessment. P.J. Edwards, J. Bembridge, M.Earl, L.Anderson, and D.Jackson. Many wildlife are dependent upon plant seeds in their diet for large parts of the year, particularly outside of the breeding season. During field trials conducted to improve risk assessment by measuring residues on food items, measurements were made to determine residues on seeds from a range of species. Seeds were sprayed either on the plant or on the ground. The effect of dehusking was investigated because many seed eating birds dehusk seeds prior to ingestion. Effects of seed size on residues were also investigated, partly by comparison with grain residues. Recovery of seed from the soil surface after application was achieved using an artificial soil surface. We conclude there is no need to discriminate between seeds sprayed on the plant or on the ground or between residues on large seeds and seeds in pods because residues are similar. There is a need to discriminate between residues on large and small seeds. Large seeds (cereal grain) = 2.7ppm fresh weight @ 1kg/ha. Small seeds (weed seeds) = 43ppm fresh weight @ 1kg/ha. There is also a need to discriminate between birds which do and do not dehusk seeds before consumption. About 80-95% of the spray residue is on the seed husk. Dehusking small seeds lowers mean residues from 43 to 5.6mg/kg (fresh weight). Small passerine birds <50g generally dehusk small seeds and would be exposed to mean residues of 5.6mg/kg in diet @ an application rate of 1kg/ha. Larger birds >50g are more likely to eat large seeds without dehusking and would be exposed to mean residues of 2.7 ppm in diet @ an application rate of 1kg/ha.

PMP037 Mechanisms to Access and Apply Scientific and Commercial Data to Address the Legal Requirements of FIFRA and the Endangered Species Act. The FIFRA Endangered Species Task Force, LLC (*FESTF*) is comprised of agrochemical companies and was formed in response to regulatory data requests generated by the Endangered Species Protection Program (*ESPP*). ESPP, within EPA's Office of Pesticide Programs (*OPP*), is charged with managing OPP's threatened and endangered species (*T&E species*) issues. The project described here is undertaken by the FESTF to meet their regulatory requirements by improving the consistency, quality, availability and use of existing information on T&E species and pesticide use. The foundation of this effort will be the development of access to existing information on T&E species, as well as a mechanism to consistently process the accessed data for use in the registration of pesticides. Consistency in the evaluation of endangered species with respect to pesticide use and impact will be achieved by providing (1) criteria for core elements that should meet minimum standards; (2) types of species data that should meet minimum data standards; (3) the relative ranking of the importance of various data elements or *fields*; and (4) minimum standards to assure a high level of data quality and accuracy.

PMP038 Effective Use of Comparative Risk Assessment to Support Expedited Review of New Reduced Risk Pesticides. Kent, D.J., The Weinberg Group, Washington, D.C. Provisions in the Food Quality Protection Act of 1996 require EPA to develop procedures and guidelines for expedited review of pesticidal active ingredients. In part because of these new requirements, and in part because of a need to address the trend toward development of more specific-action pesticides, EPA has established a Reduced Risk Initiative for conventional pesticides. New products which can demonstrate reduced risk characteristics receive expedited FIFRA registration review, and therefore are likely to reach market quicker than non-reduced risk chemicals. One of the most effective components of a successful reduced risk application is a comparative ecological risk assessment. A methodology is presented in which a new active ingredient is compared against a series of ingredients used in competing products for the same use pattern. Comparative data can be acquired using simple screening level tools such as GENEEC for the estimation of aquatic environmental concentrations or the Kenaga nomogram for terrestrial concentrations. Tools are also available for the comparison of relative mobility to groundwater. Toxicity data can be collected using general references or online databases and literature reviews. Alternatively, toxicity can be estimated through the use of Quantitative Structure Activity Relationship (QSARs) programs. The final product of the comparative risk assessment is the development of relative risk values, which are then used to establish the reduced risk of the new compound versus existing agrochemicals.

PMP039 An Automated *In Vitro* Dermal Absorption (AIVDA) Method: Human Skin Absorption of Atrazine Following Swimming/Bathing Exposure. Moody, R.P., Health Canada, Ottawa, ON, Canada. The Automated *In Vitro* Dermal Absorption (AIVDA) HPLC method that employs autosampler vial inserts to hold small viable human skin specimens (0.07 cm^2 exposed surface area) was further developed for infinite dose, swimming/bathing exposure studies. Skin absorption of the herbicide atrazine ((2-chloro-4-ethylamino-6-isopropyl-amino-1,3,5-triazine) was tested under both finite and infinite dose conditions and the data was compared to that obtained using the Bronaugh flow-through cell. The efficacy of a barrier cream to prevent atrazine skin absorption was also tested. There was no significant difference (Student's t test: $p > 0.05$) between atrazine absorption in frozen breast skin with ($23 \pm 2.5\%$ ($n = 4$)) or without ($21 \pm 4.6\%$ ($n = 3$))

barrier cream as determined by AIVDA finite dose tests. Bronaugh cell finite dose tests with fresh abdomen skin demonstrated 28 ± 7.7% (n = 4) and 28 ± 13.9% (n = 4) absorption in low (9 µg/cm^2) and high (91 µg/cm^2) dose application studies, respectively. There was no significant difference between the atrazine absorption determined by the AIVDA and Bronaugh cell finite dose tests. The infinite dose AIVDA test with fresh breast skin gave only 0.9 ± 0.02% (n = 4) and 1.4 ± 0.27% (n = 4) absorption for untreated (viable) and 1% phenol treated (nonviable) skin, respectively, this expressed as the % absorption of the total atrazine present in the pool water. Confirmation that skin viability was maintained throughout the AIVDA 24 hr infinite dose tests without phenol was obtained in a separate study by Radio-HPLC monitoring of glycolytic ^{14}C-lactic acid formation in ^{14}C-glucose spiked receiver solution. Advantages of the AIVDA method include rapid, sensitive, flexible and cost-effective analyses.

PMP040 Fingerstick Measurements of Blood Cholinesterase from California Migrant Housing Center Residents. Wilson, B.W.*; Henderson, J.D.; McCarthy, S.A.; Billitti, J.E.; McCurdy, S.A. University of California, Davis, CA. Blood cholinesterases are common biomarkers used to monitor exposure of farm workers to anticholinesterase pesticides. This study was designed to standardize and improve acetylcholinesterase (AChE) measurements of fingerstick blood from adult residents of migrant housing centers in the Central Valley of California. Fingerstick samples were collected early and late in the spray season. The blood was immediately hemolyzed with a phosphate/Triton X-100 buffer, frozen on dry ice and stored in an ultra-low temp freezer. Assays were conducted with a 96 well microplate reader using a modification of the Ellman assay at 410 nm under optimal conditions for the hydrolysis of acetylthiocholine. Mean AChE values from fingerstick and venous blood draws from 13 laboratory volunteers averaged 14.6 ± 1.2 and 14.7 ± 1.2 nmol/min/mg Hb, respectively. Preliminary results of early season AChE levels from 894 subjects were similar, averaging 14.6 ± 3.2 nmol/min/mg Hb. Late season AChE levels from 498 subjects were 12.0 ± 1.1. The results to date show reproducible blood ChE levels can be obtained from fingerstick samples if due care is taken in sampling, storage of samples, and the conditions of the assay itself. Supported in part by NIOSH (CDC U07/CCU906162-06) and NIEHS (ES05707) Center Grants.

PMP042 Passerine Response to Chlorpyrifos Exposure in Corn Agroecosystems. Richards, S.M.*, Anderson, T.A., McMurry, S.T., Hooper, M.J., Wall, S.B., Kendall, R.J. The Institute of Environmental and Human Health, Texas Tech University and Texas Tech University Health Science Center, Lubbock, TX, 79416. Chlorpyrifos [0,0-diethyl-0-(3,5,6-trichloro-2-pyridinyl) phosphorothioate] is the active ingredient in liquid and granular formulations of corn root worm insecticides, including Lorsban® 15G. Laboratory studies have shown that chlorpyrifos exerts toxicity by inhibition of cholinesterase in both the blood and brain of selected avian species. Concern exists for potential exposure of songbirds to chlorpyrifos in corn agroecosystems, which could result in mortality. A paired-plot study paradigm consisting of two cornfields near Chariton, Iowa was chosen to evaluate the potential for songbird exposure to chlorpyrifos. Lorsban® 15G was applied in a T-band according to label specifications (1.6 kg A.I./ha). Prior to and following pesticide application, representatives from 30 species of songbirds (Order Passeriformes) were caught to facilitate blood collection. Subsequent analysis of the songbird plasma was conducted for quantitation of cholinesterases. Results of cholinesterase analysis indicate that while some individual songbirds may have been exposed to chlorpyrifos, the mean level of cholinesterase activity between treatment and control populations was not significantly different ($p > 0.05$). Additional exposure-related data included soil, invertebrate, and earthworm samples at numerous locale and timepoints within the plots. Soil residue analyses from the treatment site indicated dissipation of chlorpyrifos to levels of ≤ 1ppm within 64 days of application. Low levels (≤10.6ppm) of chlorpyrifos were detected in potential bird food items (earthworms, invertebrates); however, positive detections were quite variable and thus potential exposure of chlorpyrifos to birds from these food items would also vary.

PMP043 Influence of Episodic Exposures of Chlorpyrifos to an Estuarine Cyprinid, *Fundulus heteroclitus*. Karen, D.J.*, Bielmyer, G.K., Dancik, J.A., Skipper, S.L., Smink, J.A., Klaine, S.J., Clemson University, Pendleton, S.C. Compromised bone strength, most likely a measure of structural integrity, decreased after a few short exposures to chlorpyrifos. Various theories, including biochemically and mechanically mediated processes, have been discussed in an attempt to identify the possible causes of the apparent loss of structural integrity. Impaired activity of the ultimobranchial glands, pituitary gland, and corpuscles of stannius, especially following toxic insult, may interfere with normal calcium metabolism. This may explain why biochemically-mediated pathways can weaken vertebral structural integrity. However, continual contracture of the hypaxial and epaxial musculature can weaken vertebral strength by fracturing associated bone tissue. Episodic exposure to chlorpyrifos can inhibit AChE and weaken bone strength. The primary objective of this study was to examine the effects of short term exposures to chlorpyrifos, followed by a recovery period (pulse-dosing), on *F. heteroclitus* brain AChE activity, P450 induction, and caudal vertebrae strength and composition. All parameters were measured following four 6 hour pulsed exposures to chlorpyrifos. This dosing cycle imitates natural conditions (rainfall and simultaneous runoff) observed in estuarine tidal creeks. During the test organisms were exposed to 2.5 µg¥L^{-1}, 5.0 µg¥L^{-1}, 10.0 µg¥L^{-1} of chlorpyrifos. Positive and negative controls were used to account for osmoregulatory effects. Our results demonstrate that a six hour pulsed dose inhibits brain AChE activity and does not induce P4501A1.

PMP044 Effects of Chlorpyrifos on Macroinvertebrates in Constructed Wetlands. *A. L. Bohannon,* M. T. Moore, University of Mississippi, University, MS; C. M. Cooper, USDA Sedimentation Laboratory, Oxford, MS; and J. H. Rodgers, Jr., Clemson University, Pendleton, SC. Macro invertebrate samples were collected in five different constructed wetlands, four experimental wetlands amended with chlorpyrifos and one untreated control wetland, over a two-month sampling period. Samples were collected one week prior to chlorpyrifos treatment, 48 h following chlorpyrifos treatment, and six weeks after chlorpyrifos treatment, using a Ponar grab and a D-frame kick net. Targeted individual concentrations, following a simulated rainfall event, ranged from 73 to 733 µg/L. Mean Macro invertebrate densities, total number of taxa, and percent (%) *Ephemeroptera* were calculated in each wetland on three different sampling dates, and compared using a one-way ANOVA. Significant differences in the total number of taxa were observed between some wetlands; however, no significant differences were observed in densities or % *Ephemeroptera*. Several primarily benthic taxa and one primarily planktonic taxon were collected in unamended wetlands throughout the sampling period and in all wetlands before chlorpyrifos additions. They were not collected in any treated wetlands after chlorpyrifos additions.

PMP045 A Comparison of the Toxic Effects of Two Insect Growth Regulators on the Daphnid, *Ceriodaphnia dubia*. Fetzer, B.E.*, Ramey, B.A., Eastern Kentucky University, Richmond, KY. Laboratory chronic aquatic toxicity tests were conducted to determine the relative toxicity of halofenozide and tebufenozide to the daphnid, *Ceriodaphnia dubia*. These pesticides were designed to control turf grass pests by altering the molting process. Both halofenozide and tebufenozide mimic the molting hormone 20-hydroxyecdysone and are Lepidopteran specific in their action. Since these pesticides were designed for broad terrestrial application, the potential for non-point source runoff into aquatic ecosystems poses a question as to their effect on aquatic arthropods. To address this question test animals were exposed to both commercial and technical grade compounds in standard 8-day static renewal chronic tests. Results indicate that tebufenozide is more toxic than halofenozide to *C. dubia* when both technical and commercial grades of the compounds were compared. It also appears that *C. dubia* may be more sensitive to the effects of these pesticides than other species tested.

PMP046 Aquatic Mesocosm Studies Provide Insight Into Ecological Significance of Effects of Cotton Pyrethroids. Giddings, J.M.*, Springborn Laboratories, Wareham, MA; Solomon, K.R., University of Guelph, Guelph, ON; Maund, S.J., Zeneca Agrochemicals, Jealott's Hill Research Station, Bracknell, Berkshire, UK. Results of mesocosm and field studies with cypermethrin and es-fenvalerate were analyzed and interpreted to support an ecological risk assessment of cotton pyrethroids in aquatic ecosystems. A core group of seven mesocosm studies conducted on two continents over the course of a decade were examined, and additional observations from mesocosm and field studies with these and other cotton pyrethroids were also brought to bear. The results for cypermethrin and es-fenvalerate were remarkably consistent. They revealed a trend in sensitivity from amphipods, isopods, midges, mayflies, copepods, and cladocerans (most sensitive) to fish, snails, oligochaetes, and rotifers (least sensitive). With few exceptions, populations affected by pyrethroids in the mesocosms recovered to normal levels before the end of the year of exposure; most populations recovered within weeks. Factors leading to population recovery included internal refuges (areas of low exposure), resistant life stages, rapid generation times, and egg deposition by adults from outside the treated systems. The effects of pyrethroids observed in the mesocosms were correlated with maximum pyrethroid concentrations in the water, not with concentrations in the sediments. Effects did not appear to increase with cumulative exposure from repeated pyrethroid applications. Indirect effects on fish (which have been hypothesized to occur when invertebrate food sources are reduced) were not observed. The Lowest Observed Effect Concentrations for the overall ecosystems for cypermethrin and es-fenvalerate corresponded to the 49th and 62nd percentiles of acute toxicity endpoints (LC50s) for arthropods measured in laboratory studies with these compounds, implying that a risk characterization based on 10th percentiles would be highly conservative.

PMP047 Organophosphorus insecticides and endosulfan in surface waters of the Niagara fruit belt, Ontario, Canada. Struger, J.*, Ecosystem Health Division, Environment Canada, Burlington, ON, Canada. In 1996 and 1997 surface water samples were collected from two streams in the Niagara fruit belt region of Ontario. This region supports intensive peach, grape, apple, pear, and plum production and is subjected to a large number of pesticide applications to control fruit pests. Two streams were monitored at various times during the growing season as well as outside the season for organophosphorus insecticides and endosulfan. Azinphos methyl was detected in 15/39 surface water samples (up to 2.66 ug/L). Diazinon was detected in 28/39 samples (up to 25 ug/L). Chlorpyrifos was detected in 8/39 samples (up to 0.261 ug/L). Endosulfan was detected in 32/39 samples (up to 0.243 ug/L). Other OP and OC pesticides were also occasionally detected. Detections occurred most often during periods of application and pesticides were probably entering the stream as a result of spray drift. There were also a number of water samples that contained more than one insecticide or had mixtures of organophosphorus pesticides and/or endosulfan. These results will be discussed in relation to aquatic toxicity data, pesticide use information, and possible implications to aquatic ecosystems of Ontario.

PMP048 Use and Limitations of Indices of Pesticide Mobility and Leaching Potential. Pisigan, Jr., R.A., U.S. EPA, Washington, DC. Several indices for assessing mobility and leachability of organic chemicals were examined to evaluate their uses in determining the likelihood of pesticide occurrence in groundwater. The indices were based on the models called LEACH, Attenuation Factor (AF), GUS, and those reported by California Dept. of Food and Agriculture (CDFA) and Jury and co-workers (Jury). Organic carbon sorption coefficient (Koc) and soil half-life ($t_{1/2}$) were common parameters in all models. Only AF and Jury's model took into account certain soil and hydrogeological parameters. Thiabendazole, tebuthiurun, simazine, prometon, chlorobenzilate, DCPA, dinoseb, terbacil, alachlor, carbaryl, pronamide, and chlorpyrifos were used in the indices evaluation. The pesticide properties ranged from 20 - 6000 cc/g for Koc, 10 - 360 days for $t_{1/2}$, 0.4 - 2300 mg/L for water solubility, and 0.014 E-6 - 22 E-6 mm Hg for vapor pressure. Some differences were noted in the relative leachability ranking between LEACH and AF. Pesticides predicted as leachers and nonleachers from GUS and CDFA were generally the same. Pesticides assessed from Jury's model as leachers or groundwater risks under high pollution potential scenario were tebuthiurun, simazine, prometon, DCPA, dinoseb, and terbacil. Carbaryl and pronamide, considered as nonleachers according to GUS, CDFA, and Jury's model, were detected in some groundwater wells based on a recent monitoring report. The indices can be used as a qualitative screening or ranking procedure and caution should be exercised in applying them because certain models assumptions are not likely to be perfectly met under natural conditions. Variability of Koc and $t_{1/2}$ was also a concern. The simple indices expectedly did not account for other factors such as rainfall, temperature, pesticide application rate, and preferential flow. If there was preferential flow, the distinction between leachers and nonleachers could be meaningless.

PMP049 Assessing the Environmental Impact of Vegetable Production: Surface Runoff of Pesticides and Soil from Plastic Mulch versus Vegetative Mulch. Rice, P.J.*, McConnel, L.L., Isensee, A.R., Sadeghi, A.M., Heighton, L.P., Hapeman, C.J., USDA/ARS, Beltsville, MD. Polyethylene mulch is used in vegetable production to control weeds and maintain soil moisture. Large-scale farming operations favor the use of plastic mulch despite the high inputs and excessive runoff associated with this management practice. Runoff from agriculture has been identified as a major contributor to water quality degradation and has been implicated in the failure of shellfish on Maryland's Eastern Shore. Scientists from the Beltsville Agricultural Research Center have developed an alternative vegetable production system which utilizes cover crops for mulch. Vegetative mulches have been shown to improve soil quality and eliminate the need for polyethylene. Little is known about the water dynamics and agrochemical fate within these systems. The objective of this project is to obtain quantitative data to assess the environmental impact of polyethylene and vetch mulch for vegetable production. Field studies were initiated to 1) compare the total quantity of surface runoff, 2) quantify the level of soil/sediment removed from the field in the surface runoff and 3) determine the concentrations of endosulfan, esfenvalerate, chlorthalonil, and metribuzin in the water phase and adsorbed to suspended solids of the surface runoff. Laboratory sorption and simulated rainfall studies were performed to evaluate the sorption of these agrochemicals to polyethylene and the influence of plastic mulch on their off-site movement. Data from the field experiment demonstrated that runoff from the plastic mulch is two to ten times greater in quantity than runoff from vegetative mulch. Soil erosion was reduced in the vegetative mulch plots. The information obtained from this research will be used to formulate recommendations for vegetable producers to reduce the environmental impact of vegetable production.

PMP050 Modeling the Environmental Fate of Chlorothalonil in Costa Rican Banana Plantations. Chaves-Chavarria, A.*, Shea, D., North Carolina State University, Raleigh, NC. Costa Rica is the second largest banana exporting country in the world. This high productivity is the result of a monoculture agricultural system that requires intensive use of agrochemicals. However very few studies have been conducted on the environmental fate of pesticides applied to banana plantations or under tropical conditions. Due to the limited resources available in the developing world for environmental monitoring, it is important that we develop pesticide fate models that can be used to help assess the risks of pesticides to human and ecological health. In this poster, a modeling analysis is presented for chlorothalonil use on banana plantations using a fugacity-based multimedia model. The site that we modeled is an 261 ha plantation operated by Escuela de Agricultura de la Region Tropical Humeda (EARTH) on the eastern coastal plain of Costa Rica. Chlorothalonil, a contact fungicide, is applied an average of 42 times a year by aerial application at a rate of 0.87 kg/ha. Average annual rainfall in the zone is 4000 mm and average temperature is 27 °C. Our initial model results yield a mass balance of 0.03 % in air, 1.3 % in surface water, 97.1 % in soil, and 1.6 % in sediment. Concentrations of chlorothalonil are 0.14 ng/m^3 in air, 495 ng/L in water, 0.77 ng/g in soil, and 2.89 ng/g in sediment. The estimated overall half-life of chlorothalonil at this plantation is 30 days. Comparison of predicted concentrations in the water with the acute LC50 for fish (49 ppm) suggests that chlorothalonil will not be acutely toxic. The model results are consistent with our initial field observations at the plantation.

PMP051 Modeling the Effects of Atrazine on Nitrogen Cycling in Streams: Implications for Nitrate Polluted Systems. *Laursen, A.E.* and Carlton, R.G., University of Notre Dame, Notre Dame, IN. The effects of atrazine on nitrogen cycling were modeled to assess potential impact on nitrate and ammonium concentrations. Oxygen and nitrate micro electrode data assessing response of photosynthesis, respiration, nitrification, and denitrification in sediments, were used in the model. Model results were compared with measured concentrations of nitrate and ammonium in artificial streams treated with atrazine. Nominal atrazine treatment levels were 0, 10, 50, and 100 µg L^{-1}, although measured concentrations were 0, 6.4, 25, and 43 µg L^{-1} in surface and 0,3.1, 11.3, and 19.9 µg L^{-1} in pore water by day 6. Loss is probably due to adsorption. Modeled data agreed well with experimental data, predicting decreased nitrate and increased ammonium concentrations in streams exposed to atrazine. With increased loading rates of nitrogen, streams with 50 and 100 µg L^{-1} atrazine had elevated nitrate and ammonium concentrations. Nitrate remained lower than control in streams with 10 µg L^{-1} atrazine, while the difference between control and 10 µg L^{-1} became negligible with respect to ammonium. The results suggest the most common effect of atrazine will be to reduce nitrate, and atrazine should not, therefore, exacerbate nitrate contamination in streams.

PMP052 Reduction of Nutrients in Storm Flow from a Golf Course by a Natural Riparian Wetland. Casey, R.E.*, Klaine, S.J., Clemson University, Pendleton, SC. Previously, runoff from turfgrass was sampled during natural storm events at sites above and below a wooded riparian zone. Runoff contained significant loads of nutrients (up to 6.5 kg nitrate and 10.8 kg phosphate) at the upstream site but nutrient loads were absent or significantly attenuated at the downstream site. Removal efficiencies ranged from 70 to 100% for nitrate. Phosphate was never detected at the downstream site. In order to further characterize the fate of nutrients in this system during storm events, irrigation water was amended with nitrate, phosphate and bromide and directly pumped into the riparian zone creating an artificial storm event. The fate of these chemicals was monitored by taking a time course of samples from four transects of lysimeters. The ratios of the nutrient concentrations to the conservative tracer concentration were used to determine the magnitude of attenuation. The bromide tracer demonstrated that water from surface runoff moves extensively through the subsurface of the wetland and does not follow preferential flowpaths in the subsurface. Significant attenuation occurred for both nitrate and phosphate. In addition, soil cores were taken from positions corresponding to the locations of the lysimeters. These were analyzed for organic and inorganic phosphorous and denitrification enzyme activity in order to determine whether spatial gradients existed in the wetland due to the chronic influx of nutrients from the golf course. These analyses showed that there are significant pools of both organic and inorganic phosphorous in the hydric wetland soils and that these soils also have a high capacity for denitrification. The results of this study have helped to elucidate the mechanisms by which this wetland successfully attenuates nutrient loads in turfgrass runoff.

PMP053 Potential Impacts of Forest Herbicide Applications on California's Native American Basketweavers. Currie, R.C.*, Cal/EPA Dept. of Pesticide Regulation, Sacramento, CA; Ross, J. H., Cal/EPA; Dong, M. H., Cal/EPA; Formoli, T., Cal/EPA. We used the environmental fate and exposure model CalTOX™ to estimate exposure of the granular forestry herbicide hexazinone (Pronone 10G) to Native American gatherers. A model validation with a field study compared results at 30 days post-release. Critical exposure compartment concentrations, plants and upper soil layers, compared well (Table 1). Compartment concentrations resulting from off-site movement (surface water, sediment) were not predicted accurately. Estimates of full-time, year round exposure on site yielded exposure in the 2E-5 mg/kg-day range for an exposure duration of one year. We estimate a hazard ratio of 0.0014 using a non-cancer total average daily intake of 0.033 mg/kg-d. Hexazinone does not appear to pose a significant health risk to these gatherers given this scenario. National Forest vegetation management practices impact Native American gatherers. Native Americans gather in these public lands for basket weaving materials (willow, deergrass and brackenfern) and for traditional foods (acorn and elderberry). Biological monitoring or passive dosimetry information is not available. Our indirect methods relied on environmental monitoring of plants impacted by herbicides and environmental and exposure modeling.

Table 1. Concentrations of Hexazinone in Environmental Media (Day 30)

Compartment (Day 30)	CalTOX™	Literature	Units
Air	2.6E-14	NA	mg/m^3(air)
Plants	17	11	mg/kg (fresh mass)
Ground-surface soil	19	1.1	mg/kg (total)
Root-zone soil	4.8	1.1	mg/kg (total)
Vadose-zone soil	1.4E-03	5.0E-02	mg/kg (total)
Surface water	6.3	0.035	mg/L (water)
Sediment	1.0E-04	0.75	mg/kg (sediment)

PMP054 Species decline: Contaminants and other contributing factors. Pattee, O.H., Rattner, B.A.*, Eisler, R., U.S.G.S. Patuxent Wildlife Research Center, Laurel, MD. Members of over 1,200 taxa have been listed as Threatened or Endangered, and over 4,000 additional organisms have been identified as Candidate Species or Species of Concern. Identification of critical limiting factors may result in management actions that stabilize vulnerable populations and insure their perpetuation. Both naturally-occurring and anthropogenic activities (e.g., environmental contaminants and pollution) have been demonstrated to be a significant factor in depressing populations or catalyzing the final crash of some species. The objective of this project is to develop a synthesis document and database that lists and ranks the presumed causes of decline, with special emphasis on contaminants and pollutant-related situations. This will be accomplished by synoptic review of all recovery plans (n=479) with listing packages (n=1134) serving as a secondary source of information, followed by itemization, cross-referencing, enumeration, and ranking of contributing and limiting factors. To date we have analyzed all of the recovery plans for reptiles (n=26) and amphibians (n=6). 188 causes are defined, falling into 6 major categories: habitat alteration/availability (47.8%); exploitation/harvest (19.7%); introduction of exotic species (10.1%); contaminants (9.0%); miscellaneous others (6.9%); pollution (6.4%). The applicability of these data are extensive, including facilitating reviews of Section 7 consultations and Environmental Impact Statements, reviewing permit applications, conducting environmental contaminant risk assessments, identifying specific data gaps and

research needs, selecting potential management actions, and establishing priorities for broad-based research on limiting factors applicable to groups of species rather than the current species-by-species approach. However, caution must be exercised in the use of this data because of the speculative nature of the causes; most of the causes (69.7%) are base on poorly documented expert opinion and/or guesswork. This is particularly true of the contaminant/pollution categories where only 13.8% of the incidents are documented in the literature as true causes of decline.

PMP055 An Ecotoxicological Assessment of Urban Stormwater Runoff using Laboratory and *In situ* Toxicity Testing. Moore, L.A.*, Burton, G.A., Jr., Wright State University, Dayton, OH. An ecotoxicological assessment of an urban stream was conducted to evaluate the primary stressors affecting the stream, to compare *in situ* versus laboratory toxicity testing, and to study the effects of low flows versus high flows. The approach involved toxicity testing of sediments and overlying waters at low and high flow both in the laboratory and *in situ*. Test organisms included *Chironomous tentans*, *Hyalella azteca*, *Pimephales promelas* and *Daphnia magna* exposed for 2 to 7 days. The receiving stream was characterized using physicochemical profiles, qualitatitive habitat evaluations, and rapid bioassessments of benthic macroinvertebrates. The results revealed different stressors dominated the indicator species responses. Low flow conditions were more toxic at some sites and high flow more toxic at others. The approach was also compared to recommended monitoring methods of the U.S. Environmental Protection Agency and the Ohio Environmental Protection Agency which produced differing conclusions.

PMP056 Inhibition of Growth of the Wetland Macrophyte, *Juncus effusus*, Following Exposure to Four Toxicants. Lytle, T.F., Lytle, J.S. Gulf Coast Research Laboratory, Ocean Springs, MS. In 48-500L mesocosms the wetland plant, *Juncus effusus*, was exposed to 16 possible combinations of four toxicants at environmentally realistic levels. Nominal levels were 192, 51 and 291 µg/L for atrazine, chlorpyrifos, monosodium methanearsonate respectively with methyl mercury chloride added to raise top cm sediment to 0.4 µg/g. 18 mesocosms received half-strength doses of all compounds. A repeat dose to half the mesocosms occurred on day 62. On days 1, 2, 4, 8, 16 and 32 after first dose and 2, 8 and 32 after second, plant clumps were removed and growth endpoints measured: mean shoot length, number of shoots and total length of all shoots. Multiple analysis of variance was applied to all data and indicated total shoot length provided most ambiguous results with only statistically significant effect being an overall positive growth effect by chlorpyrifos. The overall greatest effect on mean shoot length was demonstrated by atrazine. Though this effect was significant inhibition on day 2 and 4, by day 32 the greatest single effect in any treatment was significant enhancement in atrazine treated mesocosms (mean of 71cm vs 61 cm shoot length). Significant but lesser inhibitory effects were shown by the combination mercury, arsenic and chlorpyrifos and by the combination atrazine, arsenic and chlorpyrifos. Significantly longer shoots after 32 days of atrazine exposure were accompanied by significantly reduced number of shoots in these same plant clumps. On day 32 atrazine treated mesocosms contained 46 fewer shoots of *J. effusus* than those not treated. This exposure regime amply demonstrates the difficulty in extending controlled laboratory or greenhouse bioassays to mesocosm scale or field studies of multiple toxicant stressors to wetland plants.

PMP057 Use of Field-Derived Fish Tissue Residue Information for Understanding the Relevance of Mixture Toxicity. Dyer, S.D.*, White-Hull, C.E., The Procter & Gamble Company, Cincinnati, OH, Shephard, B.K., URS Greiner, Seattle, WA. Fish tissue residue data were obtained from Ohio EPA. Over 2400 samples collected throughout the state were analyzed for metals and chlorinated organics (pesticides, PCBs). Samples consisted of whole body composites as well as fillets with and without skin. Most were collected from channel catfish, carp and white sucker. Residue concentrations were converted to toxic units using ecological risk assessment Tissue Screening Concentrations (Shephard, 1996, USEPA National Sediment Bioaccumulation Conference Proceedings, Bethesda, MD) as the threshold effect concentrations for adverse effects. For chlorinated organics and all sample types, approximately 30% of the samples exceeded a TU of 1. Over 60% of whole body samples exceeded a TU of 1 and 10% a TU of 10. For metals and all sample types, 85% exceeded a TU of 1 and 5% a TU of 10. Over 75% of whole body samples exceeded a TU of 1 and 25% a TU of 10. The toxic unit estimates were compared to fish community status (e.g. Index of Biotic Integrity and raw metrics) at each respective sampling site via a GIS and subsequent correlative and multiple regression analysis. In addition, the influence of instream habitat (e.g. pools, stream gradient) on tissue toxic units and fish community status was determined.

PMP058 Utilizing a Whole Body Residue Approach to Investigate the Mechanism of Synergistic Toxicity of Atrazine and Organophosphate Insecticides to *Chironomus tentans*. Belden, J.B.*, and M.J. Lydy. Wichita State University, Wichita, Ks. Acute toxicity and whole body residues were determined for atrazine and selected organophosphate insecticides (OP's) to *Chironomus tentans*. The OP's included chlorpyrifos, methylparathion, malathion, phosdrin, and trichlorfon. Toxicants were applied individually or in binary combinations of atrazine with each OP. Standard 96 hour toxicity tests were utilized to obtain toxicity data. Body residues were evaluated by either organic extraction followed by GC/NPD or by use of radiolabeled OP's linked with liquid scintillation counting. Although atrazine was not acutely toxic by itself, binary combinations with OP's exhibited greater than additive toxicity. The tests were conducted using environmentally relevant levels of atrazine. Whole body residue data, measured for both individual toxicants and binary mixtures, indicates that the increase in toxicity may be attributable to an increase of the biotransformation rate of the OP's to more toxic metabolites.

PMP059 Analysis of Organophosphorus Mixtures using Acetylcholinesterase Inhibition and Changes in Zooplankton Abundance as Endpoints of Toxicity. George, T.K.*, University of Saskatchewan, Saskatoon, SK; Liber, K., University of Saskatchewan, Saskatoon, SK; Solomon, K.R., University of Guelph, Guelph, ON; Harris, M.L., University of Guelph, Guelph, ON. The toxicity of binary and tertiary mixtures of organophosphorus insecticides diazinon, chlorpyrifos, and azinphos-methyl was investigated in microcosm studies in Guelph, Ontario. Assessment endpoints included changes in population abundances and inhibition of acetylcholinesterase activity. The binary mixture study of diazinon and chlorpyrifos consisted of two phases. In phase I, a regression design using six concentrations ranging from 0.44 to 44.34 mg/L was used to determine toxicity thresholds. Concentrations were based on the 90th centiles of environmental concentrations. It was established that the minimal acute toxicity to zooplankton, shown at 0.44 mg/L, was suitable to assess mixture toxicity. Phase II, an ANOVA design with two mixtures of different proportions replicated three times, was used assess mixture toxicity. Each mixture, based on a chlorpyrifos equivalent concentration of 0.45 mg/L, was dominated by 80% of one pesticide and 10% of the remaining pesticide. The mixtures of phase I and II exhibited acute toxicity to Cladocera, and Copepoda, while Rotifera displayed resilience. Overall, the biological responses of Rotifera, Copepoda and Cladocera showed that the toxicity of the two mixtures of different proportions in phase II were equal. However, the mixture dominated by 80% diazinon displayed selective toxicity for the Cladocern *Simocephalus vetulus* and abundance of this population was not significantly altered. Acetylcholinesterase activity, which was measured in selected species of Cladocera, correlated with observed changes in population abundances. Overall, mixtures of diazinon and chlorpyrifos exhibit equal toxicity to zooplankton populations. Species selectivity, however, was observed. Measurements of acetylcholinesterase activity proved to be a sensitive biomarker of exposure to organophosphorus mixtures.

PMP060 Ecological Toxicity Reference Values for Petroleum Alkane Mixture Toxicity to Aquatic Biota. Shephard, B.K. and Webb, J.W., URS Greiner, Seattle, WA. A tissue residue-based approach has been developed to derive ecological toxicity reference values (TRV's) for assessing potential ecological risks to aquatic biota from petroleum alkane mixtures in surface water and sediment. The approach is based on three features of petroleum alkane toxicity. Alkanes elicit their toxic effects by narcosis. The lethal body burdens of narcotics is constant within a narrow (2 - 8 mmol/kg) range of tissue residues in aquatic biota, with chronic toxicity thresholds at about 10% of the lethal body burden. The toxicity of narcotic chemical mixtures is additive, meaning that the composition of the mixture does not drive toxicity, instead, toxicity is observed when the sum of the molar concentration of individual mixture components exceeds a critical body residue, set at 0.24 mmol/kg for alkanes. A bioaccumulation model was used to backcalculate the alkane concentration in water which results in bioaccumulation of the critical body residue. These water concentrations are the TRV's for alkanes. The approach predicts that the maximum water solubility of alkanes with a carbon chain length greater than C_{12} must be exceeded before any chronic toxicity would be observed, a prediction confirmed by the limited number of toxicity studies with alkanes. Empirically determined biota-sediment accumulation factors for a deposit feeding gastropod were used with the critical body residue to estimate sediment TRV's As petroleum toxicity at sites where alkane water solubility is exceeded is likely due to physical toxicity, not narcosis, the utility of the TRV's may be limited to sites with low levels of petroleum contamination, such as those where contaminated groundwater seeps into surface waters.

PMP061 Balanced experimental designs for multi-component mixtures: A novel fractional factorial design for screening mixture interactions. McConkey, B.J., Dixon, D.G., Greenberg, B.M. University of Waterloo, Waterloo, ON. Most experimental designs for assessing mixture interactions focus on interactions between chemicals tested in pairs. While this is very useful for determining the type of interaction, it is less applicable to estimating the toxicity due to several components within a more complicated mixture, as focusing on binary interactions alone misses the potential for group interactions. Additionally, if interactions within a group of 5 or more chemicals are to be assessed, the number of binary combinations required to test becomes very large. The proposed methodology for mixture toxicity assessment screens for binary and group interactions within a larger set of mixture components. Balanced designs have been generated for sets of up to 36 mixture components. The presented design reduces the number of experiments required to screen for mixture interactions as compared to testing all possible binary mixtures. Subsets of the mixture components are tested in groups of three or more, such that all binary combinations appear within the experimental design the same number of times. The resultant data set is compared with an assumed interaction models. Analysis of the data isolates potential binary interactions and determines if group interactions are present. The experimental design can be applied to isolate potentially synergistic compounds, or may be used to verify predictive models of toxicant interactions for larger groups of toxicants.

PMP062 Advantages of using a bioenergetics-based stressor-response model for evaluating biological significance of chemical exposure to fish. Beyers, D.W., Larval Fish Laboratory, Colorado State University, Fort Collins, CO. The magnitude of physiological stress induced by contaminant exposure was integrated with a fish bioenergetics model to estimate the biological significance of sublethal exposure under fluctuating environmental conditions. Physiological stress was quantified in terms of energy by measuring routine metabolism, food consumption, activity, and growth rates of largemouth bass (*Micropterus salmoides*) exposed to the organochlorine pesticide dieldrin. At short durations of exposure (1to4d), metabolic rate of exposed fish was depressed compared to controls, but at a longer duration (16d) metabolic rate increased as a function of concentration. Food consumption and growth rates of fish exposed for 16 d declined as dieldrin concentration increased. The response of each endpoint was consistent with predictions of the general adaptation syndrome. Energetic costs of contaminant-induced changes in metabolism and food consumption were integrated with a bioenergetics model to evaluate the biological significance of chemical exposure in a natural environment. Predictions of the integrated bioenergetics-based stressor-response model (SRM) suggest that effects of exposure of largemouth bass to ambient concentrations of dieldrin in lakes at the Rocky Mountain Arsenal National Wildlife Refuge are not biologically significant because the amount of energy lost by resident fish is inconsequential and can be easily offset by consuming more prey. Furthermore, influence of dieldrin exposure is small compared to effects of a natural stressor like water temperature. The SRM provides a general framework for integrating laboratory-derived exposure-response relationships with ecological processes to determine the biological significance of multiple stressors in a natural environment. Comparisons of relative effects of anthropogenic and natural stressors can be used to assess potential costs and benefits of alternative ecosystem management strategies.

PMP063 An individual based population model of *Daphnia magna* to extrapolate from individual to population level endpoints. Ratte, H.T.*, Dülmer, U., Hommen, U., Aachen University of Technology, Aachen, Germany. An individual based model (IBM) of *Daphnia magna* was developed on the basis of detailed life-table relations, obtained from factorial experiments at various food levels and toxicant concentrations (3,4-dichloroaniline, DCA). The model was validated by comparison of simulated population dynamics with observed one from experimental populations, in which *Daphnia* grew under both stressed and unstressed conditions. In addition, incorporating of life-table endpoint variances allows to conduct probabilistic estimation of population endpoints. We adapted the IBM to simulating effects of any toxicant. Inputs into the model are population start density, algae dosage rate, and EC50 for survival, growth and reproduction inhibition. If available, also dose-response functions can be used by the IBM. Model output comprises dose-response relationships for population endpoints such as maximum population growth rate, mean equilibrium density and mean biomass, as well as – most importantly - extinction probability (related to a specific period of time, e.g. 200 days) and recovery potential. The *Daphnia*-IBM is seen as a helpful tool for risk assessors to assess the relevance of toxicant caused life-table parameter modification for the population. Results from some example data sets will be presented.

PMP064 Estimation of Oil Toxicity Using an Additive Toxicity Model. French, D.P.*, Applied Science Associates, Narragansett, RI. The toxicity of oil has been difficult to quantify for a number of reasons: (1) Oils are mixtures of a large number of hydrocarbons, with variable toxicity and environmental fate. (2) Experimental designs, which maintain a constant exposure regime to experimental organisms, are difficult to construct. (3) Measurement of the components requires several specialized and expensive methods. (4) It is likely that only certain components are available to the organisms and causing the toxicity, complicating the interpretation of the results. Past studies have shown that the low molecular weight hydrocarbons, especially PAHs, are causing most of the toxicity to aquatic organisms. These components are volatile, making it difficult to expose organisms to constant concentrations in bioassay tests. The additive toxicity model (sum of toxic units) is used to estimate the LC50 of mixtures of chemicals with a similar mode of action. PAHs in oil are a mixture of chemicals causing narcosis (the mode of action). The additive model and available QSARs are used to estimate the acute toxicity of PAHs in oil to exposed aquatic organisms. Oil toxicity is a function of PAH content and composition in the oil. The toxicity of oils and refined products is estimated and verified with available bioassay data. The verified oil toxicity model may be used to estimate toxicity of untested oils.

PMP065 Appropriate Response Variables in Ecological Risk Assessment. Booher, J.L., Kastenberg, W.E., McKone, T.E., UC Berkeley, Berkeley, CA. The ecological risk assessment paradigm has made significant advances in the past decade. Nevertheless, a universal consensus regarding appropriate response variables for an ecosystem under threat is currently lacking. The traditional triplet approach used in risk assessment attempts to answer three questions, namely: what can go wrong, how likely is it to happen, and what are the consequences? Within this context, response variables used in ecological risk assessment measure the severity of the consequences. Response variables are thus described by any community or ecosystem process which plays a critical role in maintaining Òecosystem healthÓ, as defined by the investigator. Biologists and ecologists are continuously struggling to define appropriate response variables when considering the impacts of invading species on an ecosystem or biome. We have utilized the parallels, between the impacts of an invading species and the impacts of chemical pollutants, to compile potential response variables for the ecological risk assessment paradigm. Our compilation entails a description of the ecological theory supporting each response variable, and debates the appropriateness of each in the context of ecological risk assessment objectives. Response variables considered reflect either a community or ecosystems approach to ecology. We found that response variables stemming from the communities approach include species diversity, species abundance and composition, functional diversity, keystone species, and endangered species; response variables stemming from the ecosystems perspective include processes such as nutrient cycling, productivity, and energy transfer among trophic levels. Conclusions drawn from this analyses assert that subjectivity is inherent to the ecological risk assessment paradigm. The resources, objectives and values of the institution conducting the ecological risk assessment often dictate the appropriate response variables to be utilized.

PMP066 Plant Responses to PHC-contaminated Site Soils. Stephenson, G.L.*; Solomon, K.R University of Guelph, Guelph, ONT; Koper, N.; Middelraad, I.J.C.; ESG International Ltd., Guelph, ONT; Atkinson, G.F.; Atkinson Statistical Consulting, Ottawa, ONT; Scroggins, R.P.; Environment Canada, Ottawa, ONT. The toxicity of two PHC-contaminated site soils to eight species of plants was evaluated using whole-soil toxicity tests where the site soils were diluted with a matched, field-collected, reference soil. The exposure durations ranged from 21 to 61 d to accommodate differences in growth rates of the various test species. The six endpoints used to assess the effects of the contaminated site soils on plant growth included root and shoot length, wet mass, and dry mass. The EC50 values and their associated upper and lower confidence intervals were determined for each of the measurement endpoints using either logistic or exponential, non-linear, regression equations to model the effect of the different levels of contamination on growth. The data for the response metrics were transformed, when necessary, by applying a weighting factor to the data to address heteroscedasticity. Of the eight plant species tested, Alfalfa (*Medicago sativa*) was most sensitive to the condensate-contaminated soils and Grama Grass (*Bouteloua gracillis*) and Carrot (*Daucus carota*) were equally sensitive to the amine-contaminated soils. In general, soils contaminated with amines were more toxic to plants than those contaminated with condensates. The LC_{50}s for root length of plants ranged from 16.6 to 54.3 % contamination and from 3.6 to 15.3 % contamination for the condensate- and amine-contaminated soils, respectively. The endpoint associated with the lowest LC_{50} (2.1 % condensate) was root dry mass of Northern Wheatgrass (*Agropyron dasystachyum*); however, generally the LC_{50} values for wet and dry masses of both roots and shoots were between 2 and 6 % contamination level for both site soils.

PMP067 Use of Fertilizers as a Means of Controlling Mercury Uptake by Plants. Howell, J.P.*, Heagler, M.G., McNeese State University, Lake Charles, LA, USA. Research is presently being conducted to determine the effects of fertilizers on the uptake of mercury by a root vegetable (radish) and a leafy vegetable (spinach). Under the proper conditions, the terrestrial environment can be a very efficient mercury sink, slowing the influx of mercury into the aquatic environment. A soil's ability to adsorb mercury is determined by the organic matter content. While the soil's binding ability is a valuable physical barrier, there is a consequential risk that mercury, bound in the soil, will be absorbed by resident plants. For this reason, commercial compost and inorganic fertilizers were incorporated into a soil with a mercury concentration of 8 mg/kg to examine whether an organic fertilizer might better facilitate the immobilization of mercury and decrease the subsequent uptake of mercury by plants. The mercury concentration of the roots and leaves of both plant types were examined so that the comparisons could be drawn between the whole plants and below ground versus above ground uptake. Further research involved the collection of vegetables from area supermarkets and farmers' markets to examine the amount of mercury that a consumer may presently be exposed to on a daily basis.

PMP068 The Impact of Soil Composition on the Toxicity of Freshly-spilled Hydrocarbons. Wilson, V.J.*, EVS Environment Consultants, Vancouver, BC Canada; Landis, W.G., Western Washington University, Bellingham, WA. The availability of organic chemicals in soil is a function of their chemical properties, soil characteristics, and the time of contact between the soil and chemical. Lethal effects were determined for the earthworm, *Eisenia foetida*, exposed to different artificial soils freshly spiked with jet-fuel. Artificial soils were composed of varying proportions of sand, organic material (peat moss) and Kaolin clay. The ASTM test methodology for determining earthworm toxicity was used to develop concentration-response curves. Results indicated that the proportions of soil components impact the earthworm response to the jet-fuel toxicant. Artificial soils with high amounts of organic material were less toxic to the earthworms. Results also suggested that the clay component may impact toxicity. Artificial soils with the same composition of peat moss but with different amounts of clay exerted different toxicities on the earthworms. Past studies have suggested that sorption of organic molecules by organic material may serve to reduce the toxicity of hydrocarbons such as jet-fuel. However, for soils freshly contaminated with a hydrocarbon mixture, the clay component may be more important in determining the availability and subsequent toxicity of soil to earthworms.

PMP069 An Alternative to the O'Connor Method for Extracting Enchytraeids from Soil. Phillips, C.T.*, Checkai, R.T. and Kuperman, R.G., U.S. Army ERDEC, Aberdeen Proving Ground, MD. The O'Connor method is a commonly used technique for extracting Enchytraeids from soil. However, this method requires specialized equipment, including an extractor and a power source. An alternative method uses only a colloidal silica suspension (specific gravity 1.21) that causes invertebrates to float from soil onto the surface of the suspension. This method involves flooding a soil sample with ethanol, staining and adding the silica suspension (i.e., Ludox, Aldrich Chemical Co.). The efficiency of this Ludox Flotation Method (LFM) was compared with the O'Connor method for a fresh oak-beech forest silt loam (FSL) soil (5.9% OM) with an existing Enchytraeidae community, and for three defauned soils with contrasting organic matter content. These soils were: standard artificial (AS) soil (10% OM), Sassafras sandy loam (SSL) soil (2.3% OM) and O'Neil-Hall (OHSL) sandy loam soil (4.3% OM). Quantitation of extraction efficiency in FSL was determined by adding five adult *Enchytraeus albidus* to each sample. In the three defauned soils, we used thirty *E. albidus* worms representing three size classes (2-4, 7-9 and larger than 12 mm). Animals were placed in soil (30 g dry wt., n=10) 24 h before extraction. In the FSL, the number of enchytraeids was 82% higher and the efficiency was 58% higher with LFM ($p < 0.0001$) compared with the O'Connor method. The efficiency of recovery in the three defauned soils was significantly ($p < 0.0001$) higher in LFM for all three size classes. Results of this study show that LFM is a more efficient and less laborious compared to O'Connor method and can be used in both field surveys of Enchytraeidae community, and in ecotoxicological tests.

PMP070 Comparative sensitivity of enchytraeid and earthworm reproduction tests in soil toxicity evaluations. Kuperman, R.G.*, U.S. Army ERDEC, APG, MD; Simini, M., GEO-CENTERS, Inc., APG, MD; Phillips, C.T., U.S. Army ERDEC, APG, MD; Checkai, R.T., U.S. Army ERDEC, APG, MD. We compared sensitivity of the Enchytraeid Reproduction Test (ERT) with *Enchytraeus albidus* and the Earthworm Toxicity Test (ETT) with *Eisenia fetida* using organophosphate pesticide malathion. The ERT has several advantages over ETT, including greater ecological relevance, world-wide distribution, a short generation time of test species, and greater cost-effectiveness. Toxicity of malathion was studied in two soils with contrasting organic matter content, including standard artificial (AS) soil (10% OM) and Sassafras sandy loam (SSL) soil (2.3% OM). In the ERT test, reproducing adults were incubated for three weeks. After three weeks, adult worms were counted and soil with cocoons was incubated for an additional three weeks. Earthworm toxicity was determined using a 14-day acute survival test and a chronic 21-day cocoon production assay. Results showed that malathion had similar toxicity to *E. albidus* adults in AS and SSL with LOEC of 23.15 mg kg^{-1} in both soils. The ETT LOEC values for *E. fetida* were 50 and 60 mg kg^{-1}, respectively. Malathion was more toxic to *E. albidus* juveniles compared with adults in AS with LOEC of 7.75 mg kg^{-1} and EC_{50} of 7.6 mg kg^{-1}. The ETT chronic assay LOEC values were 12 and 21 mg kg^{-1} and EC_{50} were 16 and 20 in AS and SSL, respectively. Results of this study show that ERT is a more sensitive toxicity test and has the potential for replacing ETT in future soil toxicity testing.

PMP071 Implementation of TNRCC's Ecological Risk Assessment Guidance at a Brownfields Site. Champagne, L.F.*, TNRCC, Austin, TX; Kallus, A.S., Roy F. Weston, Inc., Houston, TX. In November of 1996, TNRCC released a draft guidance document on a three-tiered approach to conducting ecological risk assessments (ERA). The purpose of this document was to provide guidance to promote consistent and technically defensible ERAs within all of TNRCC's corrective action programs, not just Superfund. This presented quite a challenge as nearly all existing ERA guidelines/policies were designed for Superfund application. In addition, TNRCC was in the process of developing the Texas Risk Reduction Program to replace the existing risk reduction rules and any ERA methodologies had to interface with those human health components proposed in the new program. Among the first sites to attempt implementation of this guidance was a State Superfund site located along the Texas Gulf Coast. This 14-acre property had been used for wood treating and asphalting since around 1900 and was known to have elevated levels of polycyclic aromatic hydrocarbons and metals in surface soils and in sediments of an adjacent bayou. The site had been abandoned since the 1970s but remediation was planned to allow future industrial use of this brownfields property. A risk assessment was performed which initially focused on human health since the assumed industrial future use would likely have the majority of the property covered with buildings, parking areas, landscaping, and little or no native vegetation. However, recent development in the downtown area and expansion of a river walk caused the city to consider the property for recreational use, including an extension of the river walk across the property and as a park. Based on this potential use, much of the property would be vegetated and conducive to ecological receptors, thus requiring an ERA for the development of surface soil cleanup values. Although the ERA guidance allowed some procedural latitude, some difficulties were encountered during its implementation. Problems included insufficient directions regarding: where to begin (i.e., which tier), how to present the data, and when to consider risk management options. Despite these difficulties, the risk drivers were identified and ecologically-protective cleanup levels were established. The ERA guidance is currently being revised to better address these issues.

PMP072 Rapid Ecological Assessments in Support of Brownfields Redevelopment: Factors to Consider to Ensure a Streamlined Process. Fishman, B.E., Risk-Based Remedies, Beaver Falls, PA; Reinert, K.H.*, Rohm and Haas Company, Spring House, PA. Assessments tailored to supply rapid resolution of ecological issues at Brownfields redevelopment projects can significantly augment the process. An initial ecological assessment which considers the presence of critical species (including endangered or threatened species), critical habitat, and the potential for complete exposure pathways can streamline the redevelopment effort or identify limitations at the outset. If threatened or endangered species are actually present, redevelopment to anything other than improved habitat is unlikely. Many Brownfields redevelopment efforts take place in urbanized locales in which habitat is sparse. An initial assessment is useful to identify the presence of critical habitat, which can eliminate a project, or the lack of critical habitat. Documentation demonstrating a lack of these critical receptors or habitat supports the conclusion that additional ecological assessment may be unwarranted. In the absence of critical species or habitat, consideration of the potential that ecological receptors might experience adverse effects after redevelopment may be warranted; however, a decision to continue the assessment should be based on additional factors such as the types of stressors present or the presence of complete exposure pathways. In many redevelopment cases, a lack of potential completed exposure pathways at the location of interest supports earlier considerations and allows the redevelopment to proceed successfully. Streamlined processes for ecological assessment are necessary to allow accurate decision making early in the process. Failure to address ecological considerations may result in unnecessary difficulties at the later more costly stages of a redevelopment effort and potentially place ecological receptors at risk.

PMP073 Development of a Screening Ecological Risk Model. Anderson*, P.D. and Alsop, W.R. Ogden Environmental and Energy Services, Westford, MA; Walden, T. BP Oil Europe, B-1950 Kraainem, Belgium; Spence, L. Spence Environmental Engineering, Pleasanton, CA. BP has developed a computerized risk assessment model called RISC (Risk Integrated Software for Cleanup) to facilitate evaluation of potential ecological and human health risks and remediation of sites with hydrocarbons. The ecological component to be presented in this paper evaluates direct and indirect exposure pathways for several receptors exposed to surface water, sediments, and surface and subsurface soil. Potential risk associated with direct exposure to surface water is evaluated using chronic benchmarks. RISC evaluates risk from sediments by comparing chemical concentrations in sediments to benchmarks. Spatial extent of and magnitude of exceedance is also considered. Potential food chain risks are estimated by comparing a daily dose or body burden to a toxicity reference value. Within either the terrestrial or aquatic system, several receptors have been identified depending upon their class (birds, mammals, fish), dietary composition (herbivore, carnivore, omnivore), feeding range size and trophic position. Examples, including the input and output screens, will be presented to show how the model estimates potential exposures from indirect pathways and calculates toxicity quotients for receptors.

PMP074 Evaluation of Ecological Risk at Brownfields sites: The Pennsylvania Experience. Shaw, J.R., PA Department of Environmental Protection, Harrisburg, PA. The PA Department of Environmental Protection, together with the Cleanup Standards Scientific Advisory Board, developed an ecological site screening procedure to assess potential effects of contamination being remediated to the Statewide health standard under the Land Recycling Program (PA Act 2). The process became necessary when no generic standards could be developed in a timely manner to protect ecological receptors, and is designed to minimize the effort required to screen sites that do not require intensive ecological study, while maintaining the protectiveness of the Statewide health standard for those sites that do. The screen is an overlay to the Statewide health standards to provide, in a generic way, protection to ecological receptors when the human health standards may not necessarily be protective. The screen presupposes that sources of contamination will be removed and that natural attenuation will result in the reduction of the concentration and toxicity of constituents of potential ecological concern. There are criteria for no further action at various points in the evaluation, including if the area of soil or sediment contamination is less than 2 acres or 1000 square feet respectively, if only light petroleum products are present, if the site exposure pathways are eliminated by paved surfaces or buildings, and if constituents of potential ecological concern are not present and onsite gross impacts of contamination are not detected. The screening process calls for detailed ecological risk assessment only where there are indications that substantial ecological impacts are occurring, or may potentially occur in the future, after remediation. Options for mitigating detected impacts are presented that allow for flexibility in addressing substantial ecological effects detected by the screening process.

PMP075 The Influence of Land Use Patterns on Acid Volatile Sulfide and Simultaneously Extractable Metals in Sediments from Small Tidal Creeks of Charleston Harbor. *Jenkins, P.B.*, Scott, G.I, Reed, L.A., Dias, A.R., NOAA/NOS-CCEHBR, Charleston, South Carolina. Surface waters and sediments of small tidal creeks are often the first marine environments to have contact with contaminants associated with urban land use activities. Improved methods for predicting potentially toxic contaminant levels on indigenous living marine resources are needed. Previous work has shown acid volatile sulfide (AVS) is an important controlling factor in the bioavailability of the simultaneously extractable metals (SEM) cadmium, copper, lead, nickel, zinc, and mercury. The AVS approach accounts for the natural assimilative capacities of the sediment whereas more traditional environmental toxicological assessments, such as the AETA and WEA do not. Our work has indicated that cumulative extractable metals (CEM), cumulative metal concentrations obtained from microwave nitric acid digestion procedures, can be used in lieu of SEM in the AVS model with analogous predictive results. The (CEM-AVS) approach accounts for natural metal binding capacities within the sediment and may provide a more relevant estimate of *bioavailable* metal concentrations. The goal of this study was to evaluate AVS and CEM levels in sediment samples from Charleston Harbor and compare the toxicity predictions based on the (CEM-AVS) and the cumulative metal/effects range low ratio (CEM/ERL%) approaches. Sediment samples from small tidal creeks of Charleston Harbor within developed watersheds (urban, suburban, and industrial) and undeveloped watersheds (forested and wetlands) were analyzed for AVS, CEM, and grain size distribution. Both methods predicted a similar number of sites with potentially toxic metal concentrations, but interestingly, poor agreement existed between the two methods on a site by site basis. Additional work is needed to fully evaluate the applicability of each of these assessment protocals.

PMP076 Assessment of Sediments from the Santos Estuary Using the Sediment Quality Triad Approach. Abessa, D.M.S.*, University of São Paulo, São Paulo, SP, Brazil; Sousa, E.C.P.M., University of São Paulo, São Paulo, SP, Brazil; Robert Scott Carr, Texas A&M University, Corpus Christi, TX, USA, Rachid, University of São Paulo, São Paulo, SP, Brazil. The Santos estuary (Sao Paulo- Brazil) has received residues from harbor activities and industrial and urban sewage discharges. The contamination of its sediments has been historically documented. Moreover, in a recent evaluation, sediment samples from Santos estuary were toxic to the burrowing amphipod *Tiburonella viscana.* Currently, the sediments from Santos are being evaluated using the Sediment Quality Triad (SQT) approach. This study includes a 10-day solid phase toxicity testing using *T. viscana* and porewater tests on the embryolarval development of the sea urchin *Lytechinus variegatus*, chemical analysis of 6 metals, total PAHs and total organochlorines, and an assessment of the benthic infaunal community. This survey aims to evaluate the degree of degradation among 25 sites within the estuary and to identify the critical areas in need of remediation. We hope this survey will serve as a model for other sediment quality assessment studies and as a first step to providing the ecotoxicological data necessary for the establishment of sediment quality guidelines in the State of Sao Paulo. In this work, we intend to present the preliminary results from this (SQT) study.

PMP077 Field Validation of Acute and Chronic Sediment Toxicity Tests with the Estuarine Amphipod *Leptocheirus plumulosus*. McGee, B.L.*, Fisher, D.J., University of Maryland, Wye Research Education Center, Queenstown, MD. EPA standardization of the 28 d chronic sediment toxicity test with the estuarine amphipod *Leptocheirus plumulosus* is nearing completion and field validation of the method is a logical next step. To this end, complementary measures of acute (10d) and chronic (28d) toxicity with *L. plumulosus*, sediment chemistry and benthic community structure were evaluated at 11 sites in the Baltimore Harbor/Patapsco River system in Chesapeake Bay. *Leptocheirus plumulosus* are indigenous to this area providing the opportunity to compare laboratory responses with this species to viability of field populations at the test sites. Survival in 10 d exposures was the most sensitive endpoint, classifying 8 of 11 sediments as toxic. Four of these sediments also caused significant mortality in 28 d exposures; no additional toxicity was detected using sublethal endpoints (growth or reproduction). These results were not expected because studies by other researchers indicated comparable sensitivity among acute and chronic test endpoints. Potential explanations for the apparent discrepancy between this study and earlier ones include differences related to the feeding regime (algal species composition) and mode of action of the contaminants causing toxicity. There was good agreement between acute toxicity and benthic responses, as *L. plumulosus* were found only at 2 of 3 sites not exhibiting toxicity. Analysis of sediment chemistry included estimates of AVS-SEM, total metals, TOC and various classes of organic compounds. Results of organic analyses are pending. However, AVS-SEM ratios were less than one at all sites, suggesting cationic metals were not responsible for the observed toxicity.

PMP078 Development of Environment Canada's 14-Day Survival-and-Growth Test for Marine Sediments Using Laboratory-Cultured Spionid Polychaetes. Bruno J.* and Fennell, M., Environment Canada, North Vancouver, BC. Since 1992 methodology describing subchronic-toxicity testing using marine polychaetes has been in evolution. Several development-phases have investigated: species-suitability; test-duration, -temperature, -age and food-type; response in contaminated sediments; and influences of particle size and ammonia. Due to variability of field-collected organisms (size, age), cultures of *Polydora cornuta* (East Coast) and *Boccardia proboscidea* (West Coast) have been maintained at our Atlantic and Pacific regional laboratories for ongoing design of an effective, reproducible test. Recent efforts at the Pacific laboratory involved: food ration, ammonia-buildup/water-renewal schedule, and vessel size comparisons; a two-phase interlaboratory method-validation (preparation/participation, organism-supply); and species evaluations for survival-and-growth response and culturing efforts using five years of (rearing/testing) data. Significant findings include: the routinely-used 1L-jar-with-200mL-sediment setup and 5mg-food/worm/feeding were excessive for organism size; and *B. proboscidea* is less sensitive, ill-suited for detecting sublethal/growth effects, more difficult to culture and frequently provides too few test-age juveniles for setup due to lower reproduction rates and poor survival/settling. To date, efforts and commitment required (labour, time, luck?) for organism-supply and test initiation/teardown negate this bioassay from becoming a test, like that for 10-d amphipod-survival, for laboratories to routinely process numerous samples. To summarize recommendations for a less labour-intensive, more reliable and standardized microscale test: 300mL(tallform)-beaker-with-50mL-sediment (quicker/easier teardown), 2mg-food/worm/feeding, using *P. cornuta* (more sensitive, easier culturing, rare shortages) from cultures established by test users (with initial guidance from experienced Environment Canada researchers).

PMP079 Bioavailability and Toxicity of Cypermethrin in Sediments. Hamer, M.J.*, Maund, S.J., Zeneca Agrochemicals, Bracknell, UK. On behalf of THE PYRETHROID WORKING GROUP (PWG). A program of studies has been performed by The Pyrethroid Working Group to investigate the bioavailability and toxicity of the pyrethroid insecticide cypermethrin (chosen as a representative pyrethroid) in aquatic sediments. Natural sediments of 1%, 3% and 13% organic carbon (OC) content were used in the studies. Bioavailability of cypermethrin in water-sediment systems was measured by determining body burdens of water-column (*Daphnia magna*) and benthic (*Chironomus tentans*) organisms through time. Mean sediment BCFs were 0.31, 0.14 and 0.08 for *D. magna* and 0.63, 0.19 and 0.08 for *C. tentans*, in 1, 3 and 13% OC sediments, respectively. Bioavailability was inversely related to sediment OC content and was predictable within a factor of two. Effects of cypermethrin in sediment on the mortality and growth of the benthic organisms *Chironomus tentans* and *Hyalella azteca* were measured over 10 days. The 10 d median lethal sediment concentrations (LC50) were 3.6, 19 and 23 ug/kg for *H. azteca* and 14, 67 and 62 ug/kg for *C. tentans* in 1, 3 and 13% OC sediments, respectively. In sediments with 1% and 3% OC content, toxicity decreased as expected with increasing OC. At 13% OC, this relationship was not apparent, possibly due to differences in sediment characteristics. The amount of chemical predicted to be in the water phase at the LC50 or NOEC for each sediment was calculated on the basis on organic carbon partitioning (Koc) measurements from batch equilibrium studies. These values were very similar and comparable to toxicity values from water alone, suggesting that Koc is can be used to predict bioavailaibility and toxicity across different sediment types for pyrethroids.

PMP080 Implications of Gut Purging for Tissue Residues Determined in Bioaccumulation Testing of Sediment with *Lumbriculus variegatus*. Mount, D.R.*, L.P. Burkhard, U.S. EPA, Duluth, MN; T.D. Dawson, Integrated Laboratory Systems, Duluth, MN. Bioaccumulation test procedures using the oligochaete, *Lumbriculus variegatus*, have been developed as a means to evaluate the accumulation of chemicals from freshwater sediments. To avoid including chemicals associated with gut contents as part of the measured tissue residue, a 24-h period of purging in clean water after the uptake phase of the test has been recommended. While purging acts to reduce bias from gut contents, it also has the potential to introduce bias caused by depuration of chemical from tissues. In this presentation, a series of model calculations were used to assess the expected sensitivity of measured residues of nonionic organic chemicals to the presence of sediment in the gut and to varying lengths of purging. If organisms are not purged, the predicted influence of gut contents on measured residue is not large (generally <20%) when a biota-sediment accumulation factor (BSAF) of 1 was assumed. However, if BSAFs substantially less than 1 apply, then projected errors increase to 30-fold or more. To derive a better estimate of the time required for *L. variegatus* to clear the gut of sediment, a sediment purging experiment was conducted; results indicate that >98% of sediment had cleared the gut in 6 hours (half-life = 0.98 h). Based on these results and model analyses, a much shorter purging period of 6 h, rather than 24 h, is suggested as a reasonable guideline for many test applications.

PMP081 Predicting toxicity with numerical sediment quality guidelines. Long, E. R.* National Oceanic and Atmospheric Administration, Seattle, WA. Data from sediments collected from 1068 locations along the Atlantic, Pacific, and Gulf of Mexico coasts were assembled to quantify the degree to which sediment quality guidelines can correctly predict either toxicity or non-toxicity in laboratory tests. Data from acute amphipod survival tests were available for all samples and data from additional tests with sublethal endpoints were available for 437 samples. Samples in which none of the lower-range guidelines were exceeded were rarely toxic, indicating false negatives in only10% of the cases in amphipod tests. For most substances, 50% to 100% of samples were highly toxic in amphipod tests when individual mid-range guidelines were exceeded. Percentages of samples indicating toxicity increased as the numbers of guidelines exceeded increased. Data from sublethal tests often added 20-30% in predictive abilities. Recommendations are provided on uses of guidelines to identify both chemicals of concern and sites of concern.

PMP082 Cu Inhibition of Digestive Enzymes of Deposit feeders: Implications for Changing Benthic Community Structure. Chen, Z.*, Mayer, L.M., Bock, M., University of Maine, Walpole, ME; Weston, D. University of California, Berkley, CA; Self, R.F.L. Jumars, P.A., University of Washington, Seattle, WA. Contaminated Cu can be solubilized from sediments during deposit-feeding and the solubilized Cu can inhibit digestive enzymes of deposit feeders when Cu concentration reaches to a threshold level. We assess the potential impact of this Cu toxification on the community structure of deposit feeders by titrating Cu solutions into digestive fluids of various deposit and suspension feeders, followed by monitoring the activities of proteases, esterases, lipases, α- and β-glucosidases. The threshold toxic Cu concentrations varied among organisms and are largely determined by the concentrations of gut amino acids ([AA]). Enzymes from guts of high [AA] required addition of more Cu than those from guts of low [AA], because the former contained an abundance of high-energy binding sites on their AAs that prevented Cu interaction with enzyme active sites. Threshold Cu levels were similar among all enzyme species, suggesting the same inhibition mechanism. Cu was less effective at inhibiting enzymes at lower pH values, suggesting that hydrogen ion can compete with Cu ion for binding sites in gut AA. The results of this study lead to the counter-intuitive conclusion that deposit feeders with low digestive intensities (low enzyme activity, low amino acid concentration, low surfactancy) and high pH values are most vulnerable to sedimentary Cu toxicity, although they solubilize less sedimentary Cu than their counterparts with high digestive intensities and low gut pH. In general, suspension feeders and holothuroids should therefore be more susceptible to Cu contamination than deposit feeders and polychaetes, respectively. This differential response of deposit feeders to Cu toxification may be useful for predicting the shift of benthic community structures in a Cu-contaminated area.

PMP083 Chlorinated Hydrocarbons in Pine Needles: An Atmospheric Evaluation of the Eastern United States. Loganathan, B.G.*, Owen, D.A. Murray State University, Murray, KY; Sajwan, K.S. Savannah State University, Savannah, GA; Corser, J. Drexel University, Philadelphia, PA. Vegetation has recently been viewed as an important, dynamic, and active environmental compartment that influences the atmospheric transport, global turnover, and cycling of may semi-volatile organic pollutants. Pine needles have been demonstrated as a fixed-site, regenerative, annual monitoring matrix for the evaluation of local and regional distribution of lipophillic air pollutants. In this study, we determined the concentrations persistent and toxic organochlorine contaminants including polychlorinated biphenyl (PCB) congeners, hexachlorobenzene (HCB), DDT and its metabolites and chlordane compounds in surface resins of several species of pine needles in the eastern United States. Pine needle sampling was done at several locations in the state of Florida, Georgia, South Carolina, New York, Vermont, New Hampshire, Maine, Kentucky and Tennessee and included industrial, residential, undeveloped, PCB contaminated superfund site and Peregrine Falcon breeding locations. Analytical protocol began with Soxhlet extraction, silica gel column chromatography, and clean-up with sulfuric acid, followed by quantitation using gas chromatographic separation and electron capture detection. The results give credence to infer that (i) Among the various analytes measured, PCB congeners, HCB and 4,4'-DDE, were more frequently encountered. (ii) Among the various sampling sites, samples collected near superfund site, industrial locations and 'downwind' from industrial complexes showed higher concentrations (>40 ng g^{-1} dry wt.) of PCBs than the residential and undeveloped (<10 ng g^{-1} dry wt.) areas. (iii) Pine needles collected near PCB contaminated superfund site showed a characteristic congener pattern specific for that contaminated site.

PMP085 Rubidium and Cesium kinetics and tissue distributions in channel catfish (*Ictalurus punctatus*). Peters, E.L.*, Chicago State University, Chicago, IL; Schultz, I.R., Battelle Pacific Northwest Laboratories, Richland, WA; Newman, M.C., Virginia Institute of Marine Science, College of William and Mary, Gloucester Point, VA. We used a two-compartment, clearance volume-based model to examine rubidium and cesium pharmacokinetics in channel catfish (*Ictalurus punctatus*) after intravascular administration. We compared the apparent volumes of distribution in the central and peripheral compartments and the intercompartmental and whole-body clearances of both metals at 20.0°C and 27.5°C. Biological half-times of Rb were 15 to 16 d at both temperatures, but Cs biological half-times averaged 101 d and 85 d at 20.0°C and 27.5°C, respectively (5 to 7 times longer than those of Rb in the same individual). Both the intercompartmental and total body clearances of Rb were also 6 to 7 times greater than those of Cs. The apparent volumes of distribution for Rb in the central compartments were twice those of Cs and remained constant with temperature. The apparent volumes of distribution of both elements in peripheral compartments were large compared with their corresponding central compartments, and decreased by a similar extent with increased temperature. Cesium tissue to blood ratios were greatest for white muscle, with more than 85% of the Cs present in this tissue. Partitioning of Cs in peripheral tissues apparently decreased with increased temperature conditions.

PMP086 ^{137}Cs elimination by chronically-contaminated largemouth bass (*Micropterus salmoides*). Peters, E.L.*, Chicago State University, Chicago, IL; Newman, M.C., Virginia Institute of Marine Science, College of William and Mary, Gloucester Point, VA. The temperature-dependent ^{137}Cs biological half-times (T_b's) of lifetime-exposed largemouth bass (*Micropterus salmoides*) from a nuclear cooling reservoir at the U.S. Department of Energy's Savannah River Site were estimated from whole-body measurements of live fish and compared with literature records for acutely and chronically-contaminated fishes. The T_b's of the bass averaged 322 d (95% CI: 311-333 d), 225 d (95% CI: 220 to 230d), and 140 d (95% CI: 137 to 143 d) at 15, 20, and 26°C, respectively. These mean T_b's were 1.7 to 2.5 times longer than would be expected for acutely contaminated fishes, and 1.2 to 1.8 times longer than those predicted for fishes at steady-state with their environment according to recent models developed by Rowan and Rasmussen (1995). This slower elimination did not appear to result from slower elimination from skeletal muscle compared with other soft tissues, in that the muscle to whole-body ^{137}Cs concentration ratios after the elimination period were similar to those of freshly-caught bass. Our results suggested that elimination rates estimated from the terminal elimination components of acutely-dosed fishes may not reflect the elimination rates of fishes exposed to contaminants throughout their lifetime, even when care is taken to allow sufficient time for absorption of the dose.

PMP087 Dose Reconstruction and Food Web Calibration at a Radioactive Waste Pond at the INEEL. VanHorn, R.L.*, Hampton, N.L., INEEL, Idaho Falls, ID, Morris, R.C., Environmental Science and Research Foundation, Idaho Falls, ID, Barna, S.M., INEEL, Idaho Falls, ID. As a consequence of past activities, radionuclides with the potential to adversely affect ecological receptors have been released into the environment at the Idaho National Engineering and Environmental Laboratory (INEEL). Foodweb modeling was used to calculate the potential exposure of ecological receptors to contaminant concentrations in various media. The accuracy of an exposure model depends on the accuracy of the input parameter values and the validity of the model's structure (i.e., the degree to which it represents the actual relationships among parameters at the site) (EPA 1997). Site-specific field measurements of tissue residue levels (concentrations) are the most reliable means to calibrate model outputs or intermediate calculations. The Environmental Science and Research Foundation (ESRF) is tasked with evaluating the concentrations of radionuclides in biota at a warm waste pond at the INEEL. The ESRF has sponsored a number of studies resulting in the collection of radionuclide tissue concentrations in avian receptors near this pond. First, a dose reconstruction was performed using the existing tissue concentration data in these receptors to evaluate dose to each species. Second, the concentrations of radionuclides in the abiotic media were evaluated and decay corrected for use with the biotic data. The tissue concentration data were compared to known concentrations in the media to calibrate the foodweb modeling used at the INEEL. The use of this site and contaminant-specific verification of the foodweb model is critical to risk characterization and interpretation at the INEEL.

PMP088 Use of Radionuclide and Mercury Data to Evaluate Sedimentation Rates and Patterns in a Southeast Texas Estuary. Santschi, P., Allison, M., Asbill, S., Eek, A., Texas A&M University at Galveston, Galveston, Texas; Cappellino, S., Hammons, D., Parametrix Inc., Houston, Texas; McShea L., and Dobbs, C., Aluminum Company of America, Point Comfort, Texas. Radionuclide and mercury data were collected from sediment core samples in Lavaca Bay, Texas to assist in understanding historic mercury and sediment deposition patterns and sedimentation rates within the Bay. These data were ultimately used to (1) evaluate sedimentation patterns by defining vertical stratigraphy, (2) determine particle reworking (i.e., bioturbation or benthic mixing) rates, and (3) model natural recovery scenarios for possible remediation alternatives. The study was successful in measuring sedimentation rates and mixed layer depths for various areas within the Bay and understanding the effects of anthropogenic inputs of sediment to the Bay on overall deposition rates.

PMP089 Radiological Screening Values for Effects on Aquatic Biota. Jones, D.S., Oak Ridge National Laboratory*, Oak Ridge, TN. Radiological benchmarks for aquatic biota were developed for use at the U.S. Department of Energy's Oak Ridge Reservation as screening values to show the spatial extent of potential ecological effects and identify the need for additional site-specific investigation. The Point Source Dose Distribution approach was used to calculate water concentrations for selected radionuclides that result in a total dose rate of 1 rad per day to fish and invertebrates, which is the National Council on Radiation Protection and Measurements' recommended acceptable dose rate to natural populations of aquatic biota. These screening values incorporate internal and external exposures from parent isotopes and all short-lived daughter products. They also include exposures from all major α, β, and γ emissions for each isotope. Unlike exposures to chemicals, exposures to radionuclides are expressed as the dose rate received by the organism. Dose rates that account for the biological effects to the organism are additive. That is, the total dose rate is the sum of the dose rates normalized to biological effects for each radionuclide. If the total dose rate from all radionuclides in water exceeds a recommended acceptable dose rate, further analysis is needed to determine the hazards posed by radionuclides. If, however, the total dose rate falls below an acceptable dose rate, radionuclides may be eliminated from further study. *Managed by Lockheed Martin Energy Research Corp., under contract DE-AC05-96OR22464 with the U.S. Department of Energy

PMP090 Superoxide Dismutase and Radiocesium Activities in Rodents from Chernobyl-contaminated areas in Ukraine. Holloman, KH*, University of Georgia, Athens, Ga.; Dallas, CE, University of Georgia, Athens, Ga; Jagoe, CH, Savannah River Ecology Laboratory, Aiken, S.C.; Tackett, R, University of Georgia, Athens, Ga; Kind, JA, University of Georgia, Athens, Ga; Rollor, EA, University of Georgia, Athens, Ga; Chesser, RK, Savannah River Ecology Laboratory, Aiken, S.C. Three rodent species - *Clethrionomys glareolus*, *Apodemus sylvaticus*, and *Apodemus agrarius* - were collected (n = 121) from control and contaminated areas in the Ukraine. The contaminated sites were within the 10 km exclusion zone surrounding the Chernobyl Nuclear Power Plant, where a 1986 explosion released an estimated 2 x 10^{18} Bq of radioactivity into the environment. Liver and muscle samples were analyzed for superoxide dismutase activity and internal ^{137}Cs (radiocesium) activity, respectively. Superoxide dismutase activity differed significantly between control and exposed rodents for only one of the three species *(A. agrarius)*. No difference was found between species for control animals. Among the exposed animals, one species *(A. agrarius)* had a significantly lower superoxide dismutase activity than the other two species. Very high muscle radiocesium activities (up to 60,000 Bq/g) were found in contaminated animals; these were significantly higher than the controls for each species, which were at or below background. One species *(C. glareolus)* from contaminated areas had higher muscle radiocesium concentrations than the other two species. There was no significant correlation between superoxide dismutase activity and internal radiocesium activity for exposed animals for the three species. These results suggest that while internal radiocesium activities were highly elevated in animals from contaminated areas around Chernobyl, further research is needed to establish whether these activities can serve as a sensitive predictor of oxidative stress.

PTA001 Environmental Contaminants in Tissues of Four Eider Species from Alaska and Arctic Russia. J. Stout, The precipitous decline in spectacled (Somateria fischeri) and Steller's eider (Polysticta stelleri) populations compelled the US Fish and Wildlife Service to list both species as threatened under the Endangered Species Act. These declines have gone largely unexplained and have raised concerns about contaminant exposures on arctic and subarctic breeding grounds and on molting and wintering areas in the Bering Sea and Arctic Ocean. Liver and kidney tissues were collected from 99 eider carcasses including 51 common eiders (Somateria mollissima), 21 king eiders (Somateria spectabilis), 18 spectacled eiders and 7 Steller's eiders. Tissues underwent analysis for 26 organochlorine and 19 element and trace metal residues. Given that many arctic and subarctic marine organisms have been found with notable concentrations of organochlorines, we were surprised to find minimal concentrations in these eiders. However, relative to other marine birds, cadmium was elevated in some spectacled eiders, copper was elevated in common and spectacled eiders, lead was elevated in a few eiders believed to have ingested lead shot and selenium was highly elevated in spectacled eiders. Other than local anthropogenic sources of lead shot, it is unclear why these concentrations were elevated relative to other marine birds. Furthermore, it is not know whether these concentrations are part of an increasing temporal trend and/or whether eiders are able to tolerate such levels of elements and trace metals.

PTA002 The Effects of Fire Control Chemicals on the Hatchability of Northern Bobwhite Quail (*Colinus virginianus*) and Red-winged Blackbirds (*Agelaius phoeniceus*). Buscemi, D.M.*, University of Maryland College Park, College Park, MD; Vyas, N., Patuxent Wildlife Research Center, Laurel, MD; Hoffman, D.J., Patuxent Wildlife Research Center, Laurel, MD; Spann, J., Patuxent Wildlife Research Center, Laurel, MD; Kuenzel, W.J., University of Maryland College Park, College Park, MD. Avian eggs found in peripheral areas serving as fire breaks may be sprayed with fire control chemicals, therefore acute toxicity tests were conducted immersing northern bobwhite quail (*Colinus virginianus*) eggs and red-winged blackbird (*Agelaius phoeniceus*) eggs for 10s in different water-based formulations of Silv-Ex, a fire suppressant chemical, and Phos-Chek D-75F, a fire retardant chemical. Blackbird eggs were treated with levels of Silv-Ex ranging from 5 g/l to 100 g/l , and levels of Phos-Chek D-75F ranging from 0.07 kg/l to 0.95 kg/l. Silv-Ex, 10 g/l and 100 g/l, and 0.43 kg/l , 0.70 kg/l, and 0.95 kg/l Phos-Chek D-75F, significantly increased embryonic mortality according to the Fisher's Exact Test ($P<0.05$). Quail eggs were exposed to 10 g/l and 100 g/l Silv-Ex, and levels of Phos-Chek D-75F ranging from 0.13 kg/l to 1.21 kg/l. Curiously, lower concentrations of Phos-Chek, from 0.20 kg/l to 0.45 kg/l, and higher concentrations, 0.95 kg/l and 1.21kg/l, caused a significant increase in mortality. In addition, one of these treatment groups showed levels of plasma alanine aminotransferase that was significantly higher than the control group, and two others showed a significant elevation in plasma aspartate aminotransferase. Fire control agencies should give serious consideration to the difficult choice of the damage a specific fire is causing and the harmful effects certain formulations of Silv-Ex and Phos-Chek D-75F will cause to avian species, especially during their breeding season when embryos are developing within fertilized eggs.

PTA003 A New Approach for Canadian Tissue Residue Guidelines for the Protection of Wildlife Consumers of Aquatic Biota: A Case Study of Polychlorinated Biphenyls (PCBs). Roe, S.L.*, Caux, P.-Y., Kent, R.A., Environment Canada, Hull, PQ, Canada. The purpose of the Canadian Tissue Residue Guideline (TRG) is to provide a benchmark level of PCBs in aquatic life above which wildlife predators may be at risk of adverse effects. Due to the complex nature of PCBs, a new approach in the national guideline derivation and expression was explored. To accommodate differences in concentration and toxicity among individual congeners and between species, the TRG for PCBs was developed using the lastest dioxin toxic equivalency factors (TEFs) for mammalian and avian species. This is the first Canadian TRG which attempts to deal with the combined effects of mixtures. Moreover, it is recommended that TRGs for PCBs and Dioxins/Furans be used concurrently, given the shared mode of action for many congeners. The final guideline value is currently under review and will be presented.

PTA004 Extrapolating Toxicity Reference Values in Terrestrial and Semi-aquatic Wildlife Species Using Uncertainty Factors. Hoff, D.J.*, and Henningsen G.M. US EPA, Region 8, Superfund Technical Support, Denver, CO. A fundamental component in ecological risk assessments is the determination of xenobiotic doses to site-specific ecological receptors leading to scientifically defensible no-observable-adverse-effects-levels (NOAEL) and low-observable-adverse-effects-levels (LOAEL) of population sustainability. Unfortunately, wildlife literature toxicity data is not always available for most compounds and extrapolations of toxic doses must be done across species. Three principle techniques have been used for interspecific extrapolation of toxic responses to xenobiotics in wildlife species: uncertainty factors, scaling factors and physiologically based toxicokinetic models (PBTK). Currently, much debate is occurring as to the most appropriate scientific methodologies and national working groups are attempting to reach scientific consensus on the topic. In the interim, Region 8 USEPA has proposed an uncertainty factor scheme for interspecific wildlife toxicity extrapolation. The use of uncertainty factors and its current advantages over the other methods are discussed in this paper with specific applications of interspecific extrapolations of heavy metals. Four sources of uncertainty are quantified in the extrapolation: taxonomic relationship, study duration, study endpoint, and site-specific modifications. This method provides the skeletal structure for extrapolating and normalizing the most scientifically defensible, applicable toxicological study to a receptor of concern.

PTA005 Allometric Models for Inter-species Extrapolation of Wildlife Toxicity Data: Expanding the Database. Sample, B.E.*, Arenal, C.A., Oak Ridge National Laboratory, Oak Ridge, TN. In wildlife risk assessment, it is necessary to extrapolate toxicity data from test species to wildlife species because toxicity data are not available for all possible wildlife endpoint species. Allometric scaling, one of the available extrapolation methods, is based on the premise that excretion and metabolism of toxic chemicals are a function of an animals metabolic rate, which varies as a function of body weight$^{3/4}$. Previous research has indicated that acute mammalian toxicity data scales to body weight$^{3/4}$, while acute avian toxicity data scales to body weight^{1}. However, because these relationships were derived based primarily on data from drugs, or carbamate or organophosphate pesticides, their applicability to metals and chlorinated hydrocarbons commonly found at contaminated sites is uncertain. To address this issue, we obtained avian and mammalian acute toxicity data for over 100 chemicals for which multiple species had been tested. Mean body weight data for all test species were obtained from the published sources. Lethal dose per animal (LD_{50} x mean body weight) was calculated for each chemical and test species. Linear regression models of log-transformed lethal dose per animal on log-transformed body weight were developed for each chemical. Body weight scaling factors (e.g., regression slopes) were found to vary widely across chemicals for both birds and mammals. In general, preliminary analyses suggest that the body weight$^{3/4}$ scaling factor may not be appropriate for extrapolation of acute mammalian toxicity data. Whereas the body weight^{1} scaling factor may be suitable for acute avian toxicity data. However, due to the wide variability of scaling factors among chemicals, use of chemical-specific scaling may be more appropriate than one general scaling factor.

PTA006 Use of a Productivity Index in Assessing Agricultural Pesticide and Tillage Impacts on Songbirds of Conservation Concern. Martin, P.A.*, CWS, Burlington, ON; Dale, B.C., CWS, Edmonton, AB. The grasslands of prairie Canada are home to several endemic songbird species, some of which are suffering population declines due to the impacts of extensive agricultural development on habitat. The Baird's sparrow (*Ammodramus bairdii*) is one such species. In two large scale studies, one studying the effects if grasshopper insecticides, the other on tillage management systems on grassland songbird abundance and productivity in Alberta (1993-1996), we were able to assess reproductive success of Baird's sparrows, present in low numbers, through the use of a productivity index utilizing behavioral observations made during 5-7 visits per plot. We compared productivity values determined through this index to values obtained through point-counts, the method typically used in assessing breeding bird abundance, from which assessments of species conservation status are often made. Sparrows occurred routinely on cultivated farmland and were detected through point-counts, but had extremely low reproductive output, indicating that cultivated fields were inappropriate sink habitats. However, on native grassland either unsprayed or sprayed for grasshopper control, Baird's sparrows were able to successfully produce young. These results corroborate those found for a more common species in our plots, whose nest success we were able to monitor directly.

PTA007 Quantifying Plasma Lipids in Avians using Gravimetric, Colorimetric and Enzymatic Techniques. Drouillard, K.G.*, Trent University, Peterborough, ON, Canada, Sandau, C.D., Carleton University, Ottawa, ON, Canada, Norstrom, R.J., Canadian Wildlife Service, Hull, PQ, Canada. Quantifying lipid concentrations in blood tissues and blood plasma are important for extrapolating contaminant burdens from blood samples using equilibrium partitioning concepts. This is particularly true in samples collected from the field where plasma lipid concentrations within a species can vary markedly depending on the duration since last feeding. This study compared gravimetric, colorimetric and enzymatic methods for determination of lipids in pooled plasma samples from chickens and polar bears. Individual gravimetric trials were found to be highly variable and dependent on the solvent strength and volume of protein denaturant employed. Individual gravimetric trials of varying solvent strengths yielded lipid contents ranging from ND to 1.09% and from ND - 3.77% lipid by weight in pooled samples from polar bears and non-fasted chickens, respectively. The optimized method for gravimetric extraction utilized hexane:MtBE:MeOH: plasma (1:1:1:1) and produced lipid concentrations which compared favorably with enzymatic and colorimetric techniques (average lipid content of 1.07±0.04% in polar bear pools and 4.19% in plasma from non-fasted chickens). Colorimetric and enzymatic methods were also employed to determine plasma lipid contents in samples collected from the field in several avian species including herring gulls, bald eagles, great blue herons, red-tailed hawks and ring doves. Results indicate that gravimetric techniques in which the extraction solvent has been optimized for contaminant recoveries may underestimate the total lipid content in plasma samples.

PTA008 Monitoring Wading Bird Exposure to Agricultural Chemicals Using Serum Cholinesterase Activity. Parsons, K.C, Manomet Center for Conservation Sciences, Manomet, MA; Matz, A.C.*, Manomet Center for Conservation Sciences, Manomet, MA; Hooper, M.J., The Institute of Environmental and Human Health, Texas Tech University, Lubbock, TX; Pokras, M.A., Tufts University School of Veterinary Medicine Wildlife Clinic, North Grafton, MA. We have monitored breeding colonies of wading birds (herons, egrets, ibises) on the northeastern Atlantic coast of North America since 1986. Flightline analyses, prey item identification, and foraging observations indicated that certain species were foraging in habitats with potential for exposure to cholinesterase-inhibiting compounds. We tested for differences among colonies (in Boston Harbor, New York Harbor, Cape Cod, and Delaware Bay) and species black-crowned night- heron, cattle egret, snowy egret, little blue heron and glossy ibis) in serum total, acetyl- and butyryl-cholinesterase activities. Significant differences among colonies were, in some cases, consistent with surrounding land use patterns, including agriculture. Differences among species reflected taxonomic relationships and foraging patterns. For example, only glossy ibis showed the age-related patterns of cholinesterase activity observed in other altricial species. Also, cattle egrets had low cholinesterase activity, and foraged in upland, often agricultural, areas. Glossy ibis foraged in shallow salt marsh areas, relatively far downstream from sources of agricultural chemicals, and had high cholinesterase activity. Cholinesterase reactivation assays were positive in from 0-70% of serum samples. The lack of an expected developmental increase in cholinesterase in most species, and reactivation of samples in non-urban estuaries suggest some species may be exposed to cholinesterase-inhibiting compounds throughout the nestling period. Serum cholinesterase activities are useful for monitoring exposure to cholinesterase-inhibiting compounds in humans and livestock; widespread monitoring efforts of exposed and unexposed populations of wild birds can provide a reference database for sub-lethal exposures.

PTA009 Chlordane Poisoning of Birds in New Jersey. Stansley, W.*, Roscoe, D.E. New Jersey Division of Fish, Game and Wildlife, Lebanon, NJ. During a 16 month period in 1996-97 we documented chlordane poisoning in 6 species of songbirds and 4 species of raptors in New Jersey. Particularly noteworthy among these cases are recurring mass mortalities of birds at suburban roosts. At one roost we recovered 428 dead or sick birds (307 common grackles (*Quiscalus quiscula*), 104 European starlings (*Sturnus vulgaris*), 14 American robins (*Turdus migratorius*) and 3 unidentified birds) over a three week period in July 1997. Sick birds displayed characteristic cyclodiene poisoning symptoms including convulsions, opisthotonos and excessive vocalization. Brain metabolite residues indicative of chlordane poisoning were found in all of the 23 specimens analyzed. We believe this to be the largest chlordane poisoning incident reported in the United States. Chlordane poisoning was also diagnosed in nine Cooper's hawks (*Accipiter cooperi*), a state-listed endangered species. The timing of the Cooper's hawk mortalities coincides closely with the July peak in songbird mortalities, suggesting that the hawks may be feeding on birds debilitated by chlordane. The results of this and other studies suggest that chlordane poisoning of birds may be more common than is currently recognized.

PTA010 Organochlorine Distribution among the Chorioallantoic Membrane, Embryo and Remaining Egg Contents of Developing White Leghorn Chicken Embryos (*Gallus domesticus*). [1]Bargar, T.A., [2]Cobb, G.P., [1]The Department of Environmental Toxicology, Clemson University, Pendleton, SC, 29670. [2]The Institute of Human and Environmental Health, Department of Biological Sciences, Texas Tech University, Lubbock, TX, 79409. The distribution of PCB congeners 105, 156, 189 and endosulfan mixed isomers in developing chick embryos was investigated. Two adult hens were subcutaneously injected with 2.0 ug/ul of a chemical mixture containing the above chemicals once every four days during the experiment. Hens were artificially inseminated following one week of dosing. A total of 21 eggs were collected from each hen after insemination initiation and incubated. Six of the 21 eggs were inviable. Five of the remaining 15 were sacrificed at 9 days post of development, 5 more at 14 days of development and the remaining 5 at 19 days of development. The embryo, chorioallantoic membrane (CAM), and remaining egg contents (yolk, yolk sac, albumin) were separated from each other at the time of sacrifice, extracted and analyzed for each of the injected compounds. Comparisons were made among identical tissues at different points of development to determine the pattern of organochlorine partitioning during embryonic development.

PTA011 Organochlorine Contaminant Exposure and Effects in Pipping Black-Crowned Night-Herons in Delaware Bay. Rattner, B.A.*, Hoffman, D.J., Melancon, M.J. and Olsen, G.H., USGS Patuxent Wildlife Research Center, Laurel, MD; Parson, K.C., and Schmidt, S.R., Manomet Center for Conservation Sciences, Manomet, MA. Pea Patch Island in Delaware Bay is the site of the largest heronry north of Florida. From 1989-93, the population of 9 species of wading birds numbered approximately 12,000 pairs, but has recently declined to 7,000 pairs. Because Delaware Bay is a major shipping channel, and receives anthropogenic releases of toxic substances from agricultural, industrial and municipal point and nonpoint sources, contaminant exposure and effects to the heronry have been an ongoing concern. In 1997, piping black-crowned night-herons (BCNHs) wee collected from separate nests at Pea Patch Island (N=15), and from a coastal reference site, Middle Island (N=9), in Rehoboth Bay, DE. There was neither evidence of malformations nor hepatic histopathological lesions, and body and liver weights did not differ between sites. Biomarkers of petroleum hydrocarbons, polyhalogenated contaminant and metal exposure (cytochrome P450 induction and oxidative stress responses) did not differ ($P>0.05$)between sites, however, activities of benzyloxy- and ethoxyresorufin-O-dealkylase were greater in 3 of 15 embryos from Pea Patch Island compared to Middle Island. Concentrations of 21 organochlorine pesticides and metabolites were relatively low at both sites, *with p,p'*DDE values well below the threshold associated with eggshell thinning. Although total PCB concentration was modestly elevated ($P<0.05$) in Pea Patch Island BCNH embryos, levels of axylhydrocarbon-receptor active PCB congeners, dioxins, dibenzofurans and Toxic Equivalents were low and did not differ between sites. Surprisingly, organochlorine contaminant exposure and effects in Delaware Bay BCNHs appear to be considerably less than that observed and recently reported (ETC 16:2315-2322, 1997) for herons residing in the Chesapeake Bay.

PTA012 Quantifying Avian Mortality Resulting from Organochlorine Exposure on the Rocky Mountain Arsenal National Wildlife Refuge, Colorado. Johnson, G.D.*, T.L. McDonald, M.D. Strickland, and L.L. McDonald. Western EcoSystems Technology, Inc., Cheyenne, WY. Rocky Mountain Arsenal (RMA) was used for production of insecticides, including several organochlorines, from 1946 to 1982. Numerous dead birds have been found where dieldrin or endrin exposure were implicated as the cause of death; however, studies have not previously been conducted to quantify avian mortality. Objectives of this study were to (1) estimate total number of bird deaths attributed to organochlorine exposure on RMA, (2) estimate spatial distribution and an index to the level of avian mortality across RMA, (3) relate level of avian mortality to habitat type, distance from organochlorine point sources, and mean organochlorine concentrations where carcasses are found, and (4) establish standardized carcass search sites and methods for long-term monitoring of avian mortality on RMA. RMA is 27 square miles in size, and is largely undeveloped, open grassland. Carcass searches were conducted on two study areas (strata) based on anticipated level of organochlorines in the environment. A rectangular grid of 100mX100m sampling plots was established on each study (total=110) area using a systematic design with a random start. A systematic sample of 26 specialized habitat units identified as potential high bird use areas also was selected for sampling. Carcass searches were conducted on half the plots each week. Unique statistical methods used to estimate total avian mortality ($\pm$ 90% CI) by season and size class of bird based on mean number of carcasses found per plot ($\pm$ standard error [SE]) adjusted for mean length of stay ($\pm$ SE) (scavenging) and mean searcher efficiency ($\pm$ SE) are described in detail, as are statistical and GIS methods used to relate avian mortality level to habitat, distance to point sources, and organochlorine concentrations in the environment.

PTA013 Nestling Tree Swallow (*Tachycineta bicolor*) PCB Body Burdens and Their Effects on Reproduction and Growth. Yorks, A.L.*, Melancon, M.J., and Hoffman, D.J., USGS Patuxent Wildlife Research Center, Laurel, MD; Henshel, D.S., Indiana University, Bloomington, IN; Sparks, D.W., U.S. Fish and Wildlife Service, Bloomington, IN. Tree swallows (*Tachycineta bicolor*) were monitored during three consecutive breeding seasons at eight sites in Maryland, Pennsylvania, and New York representing a range of polychlorinated biphenyl (PCB) contamination in order to evaluate if there were any effects on reproductive or growth parameters. Eggs were collected and analyzed to determine contamination due to the females' body burden and establish a baseline PCB level. Composite sediment samples and pooled stomach contents were also analyzed to characterize each site and quantify PCBs from local food sources. Reproductive success was assessed by clutch size, and embryo and nestling survival. Clutch sizes at the two most contaminated sites near Philadelphia PA were generally smaller than at other sites. Nine-day nestlings were ligatured and insects in the food samples were identified to order (suborder for Dipterans), categorized as aquatic or nonaquatic, and analyzed for total PCBs. Growth parameters examined for twelve-day nestlings included body, liver, and heart weights, and bone, culmen, and brain symmetry measurements. A subset of carcasses were cleared and skeletons were stained in order to observe growth and bone structure. Body weights at two contaminated sites were lower than those at control or less contaminated sites. There were no consistent differences between liver and heart weights that related to contamination. While there were only subtle differences between reproductive and growth parameters from nestlings at various sites, body burdens of PCBs correlated strongly with levels in sediment, eggs, and food sources.

PTA014 *In situ* behavioral response of Common Loons to elevated blood mercury levels. Nocera, J.J., ACWERN, Acadia University, Wolfville, NS, Canada; Taylor, P.D., ACWERN, Acadia University, Wolfville, NS, Canada; Burgess, N.M.*, Canadian Wildlife Service, Sackville, NB, Canada. Common loons (*Gavia immer*) in Kejimkujik National Park (Nova Scotia, Canada) have the highest total blood mercury (Hg) concentrations of any loon population tested in North America. Also, reproductive success at this remote site is comparatively low. We hypothesized that if total blood Hg levels of Kejimkujik loons were impairing productivity, that such effects might be expressed through subtle, altered behaviors of breeding adults or chicks. We addressed this question through a comparative behavioral investigation of breeding, non-breeding, and failed breeding loons across a range of lakes that exhibit variation in lake morphometry, lake chemistry and physical characteristics in Kejimkujik and the Lepreau Crown Lands (New Brunswick, Canada). Time-activity budgets and event rates of loon behaviors on these lakes were collected and analyzed against 37 environmental variables using MANOVA. We report a significant association of sub-lethal blood Hg contamination with behavioral changes in young loon chicks. Chicks with higher blood Hg concentrations spend less time brooding (by back-riding) ($p = 0.004$) and more time preening ($p = 0.003$) than chicks with lower blood Hg levels. The sum increase in energy expenditure is not compensated for by any increase in chick feeding rates or begging. We suggest that such altered behavior may disrupt the energetic balance of developing young. Our results show that decreases in time spent back-riding are associated with decreases in fledging rate. We confirm the hypothesis that loons and other fish-eating predators could be at risk from exposure to elevated levels of bioavailable Hg in acidic lakes. This may help explain the chronically low loon productivity of such seemingly pristine sites as Kejimkujik, and allow for more focused research and management initiatives.

PTA015 Potential Risk to Wood Storks (*Mycteria americana*) From Mercury in Carolina Bays on the Savannah River Site. Brant, H.A.,Jagoe, C.H., Bryan, A.L. Jr., Gariboldi, J.C. University of Georgia, Savannah River Ecology Laboratory, Aiken, SC. Carolina bays are freshwater wetlands, the only natural lentic systems on the Atlantic coastal plain in the Carolinas. They serve as important breeding and feeding habitats for wildlife, including some endangered species such as Wood Storks (*Mycteria americana*). Bays tend to be acidic and rich in DOC, factors that favor mercury methylation and bioaccumulation. Water levels in these wetlands typically decrease during the summer months, concentrating prey and attracting storks. To assess potential risks to Wood Storks from mercury in fish consumed in these bays, we selected nine bays on the Savannah River Site (SRS), a federal reservation in South Carolina, with records of use by Wood Storks. Fish and amphibian species known to be consumed by storks were collected from these bays using minnow traps and hoop nets from March through June of 1997, yielding a total of 297 fish and amphibians from 9 different species. Samples were acid digested and analyzed for total mercury by cold vapor atomic fluorescence (CVAF). Mean mercury concentrations for each species of prey ranged from 0.01ug Hg/g wet mass in siren to 0.22 ug Hg/g in warmouth. Mercury concentrations differed significantly among species and among bays. Mercury methylation and bioavailability in these systems appears related to a suite of factors, including bay water chemistry and hydroperiod. Risk factors were calculated using two different published LOAEC values for bird prey items. These calculations suggest that there is potential concern for Wood Storks foraging in some bays, even though there is no obvious local source of mercury to these wetlands.

PTA016 Toxicity of Lead-Contaminated Sediment to Mute Swans. Day, D.D.*, Beyer, W.N., Hoffman, D.J., USGS Patuxent Wildlife Research Center, Laurel, MD; Sileo, L., USGS National Wildlife Health Research Center, Madison, WI; Audet, D.J., USFWS, Spokane, WA; Ottinger, M.A., University of Maryland, College Park, MD. The role of incidental ingestion of sediment-bound contaminants has not been adequately appreciated in assessments of waterfowl exposed to heavy metals. In this study, we examined the toxicity of lead-contaminated sediment from the Coeur d'Alene River Basin in Idaho to captive mute swans (*Cygnus olor*). A commercial avian maintenance diet combined with 0, 12, or 24% lead-contaminated sediment (with 3950 $\mu g/g$ lead) or 24% uncontaminated sediment (with 9.7 $\mu g/g$ lead) or, alternatively, ground rice combined with 0 or 24% lead-contaminated sediment or 24% uncontaminated sediment was fed to swans in groups of eight for 42 days. Swans fed the ground rice diet containing 24% lead-contaminated sediment were the most severely affected, experiencing a 24% decrease in mean body weight, including three birds that became severely emaciated. This group also had the greatest mean concentration of lead in blood (3.2 $\mu g/g$) and liver (8.5 $\mu g/g$) and had significant reductions in mean hematocrit and hemoglobin concentrations. All swans in the three groups fed lead-contaminated diets had substantial increases in protoporphyrin concentrations, while ALAD levels decreased by more than 95%. All swans fed diets that contained 24% lead-contaminated sediment had renal acid-fast intranuclear inclusion bodies which are diagnostic of lead poisoning in waterfowl. Body weight and hematological measurements of swans on control and reference sediment diets remained unchanged. Lead poisoning was more pronounced when swans were fed a rice diet than when fed a commercial diet high in calcium and protein. We concluded, therefore, that when mute swans consumed environmentally relevant concentrations of lead-contaminated sediments, they were exposed to sufficient amounts of biologically available lead to develop lesions and hematological changes indicative of severe lead poisoning.

PTA017 Bioaccumulation of Manganese in Feral Pigeons Exposed to High Concentrations of Manganese Oxide Dust (Mn_3O_4). Zayed, J., Akoume, M.Y., Sierra, P., Gareau, L., Talbot D., Université de Montréal, and Kennedy, G., École Polytechnique, Montréal (Québec), Canada. Birds have been used previously as biological indicators of urban lead pollution, demonstrating differences among areas of heavy and light traffic density. The present study is a step in considering the potential of feral pigeons (*Columba livia*) for monitoring manganese (Mn) contamination. We have examined the bioaccumulation of Mn in various organs/tissues and feces of pigeons exposed to very high levels of Mn_3O_4 via inhalation. A group of 4 pigeons (E^+) was exposed to 11,000 µg m^{-3} for 6 hrs/day, 5 days/week for 3 consecutive weeks and compared to a control group (E^- n=4) exposed to the backgroud level concentration of 0.02 µg m^{-3}. Mn contents in water, food, tissues, organs and feces were determined by neutron activation analysis. Results show significant differences ($p<0.05$) between E^+ and E^- for Mn blood (147.2 ppb vs 26.9 ppb), lungs (37.4 vs 0.2 ppm), trachea (10.1 vs 0.6 ppm), kidney (10.4 vs 5.8 ppm), liver (6.2 vs 1.8 ppm), brain (0.88 vs 0.49 ppm), pancreas (3.9 vs 1.2 ppm), femur (8.2 vs 1.6 ppm) and feces (111 vs 24 ppm). However, intestine and wishbone do not show any significant difference. At the end of exposure period, the exposed group seemed to be shapeless compared to the control group. These results indicate that at very high levels of exposure almost all tissues and organs accumulate Mn and thus have a potential for monitoring Mn contamination.

PTA018 Toxicity of Lead-Contaminated Sediment and Nutritional Interaction in Mallard Ducklings. Hoffman, D.J.*, Heinz, G.H., USGS Patuxent Wildlife Research Center, Laurel, MD; Sileo, L., USGS National Wildlife Health Research Center, Madison, WI; Audet, D.J., Campbell, J.K., LeCaptain, L.J., USFWS, Spokane, WA. The toxicity of lead-contaminated sediment, related to mining activity in the Coeur d'Alene River Basin (CDARB) in Idaho, was examined on posthatching development of mallards (*Anas platyrhynchos*) for 6 weeks. Day-old ducklings received untreated control diet, clean sediment (24%) supplemented control diet, or CDARB sediment (3449 ug/g lead) supplemented diets at 12% or 24%. The 12% CDARB diet resulted in a geometric mean blood lead concentration of 1.41 ppm (ww) with over 90% depression of red blood cell ALAD activity and over three-fold elevation of free erythrocyte protoporphyrin concentration. The 24% CDARB diet resulted in blood lead of 2.56 ppm with over six-fold elevation of protoporphyrin. In this group the liver lead concentration was 7.92 ppm (ww) and there was a 40% increase in hepatic reduced glutathione concentration. The kidney lead concentration in this group was 7.97 ppm and acid fast inclusion bodies were present in the kidneys of 4 of 9 ducklings. When ducklings were on a less than optimal diet (two thirds corn and one third standard diet), CDARB sediment was more toxic; blood lead levels were higher, body growth and liver biochemistry (TBARS) were more affected, and prevalence of acid fast inclusion bodies increased. Lead from CDARB sediment accumulated more readily in duckling blood and liver than reported in goslings, but at given concentrations was generally less toxic to ducklings. These findings confirm the toxicity of this exposure route as reported in wild geese and mallards within the CDARB.

PTA019 Toxicity of Lead-Contaminated Sediment to Goslings. Hoffman, D.J.*, Heinz, G.H., Obrecht, H.H., USGS Patuxent Wildlife Research Center, Laurel, MD; Sileo, L., USGS National Wildlife Health Research Center, Madison, WI; Audet, D.J., Campbell, J.K., LeCaptain, L.J., USFWS, Spokane, WA. The toxicity of lead-contaminated sediment, related to past mining activity in the Coeur d'Alene River Basin (CDARB) in Idaho, was examined on posthatching development of Canada geese (*Branta canadensis*) for 6 weeks. Day-old goslings received an untreated control diet, a clean sediment (48%) supplemented control diet, or CDARB sediment (3449 ug/g lead) supplemented diets at 12%, 24%, or 48%. The 12% CDARB diet resulted in a geometric mean blood lead concentration of 0.68 ppm (ww), greater than 90% depression of red blood cell ALAD activity, and over four-fold elevation of free erythrocyte protoporphyrin concentration. The 24% CDARB diet resulted in blood lead of 1.61 ppm, decreased hematocrit, hemoglobin, and plasma protein in addition to the above effects. The 48% CDARB diet resulted in blood lead of 2.52 ppm with 22% mortality, decreased growth and elevated plasma LDH-L activity. In this group the liver lead concentration was 6.57 ppm (ww) with two-fold increases in hepatic lipid peroxidation (TBARS) and glutathione concentration, elevated glutathione reductase activity, and lower protein-bound thiols concentration and G-6-PDH activity. The kidney lead concentration in this group was 14.93 ppm with subacute renal tubular nephrosis in one of the surviving goslings. Lead from CDARB sediment accumulated less readily in gosling blood and tissues than reported in ducklings, but at given concentrations was generally more toxic to goslings. These findings confirm the toxicity of this exposure route as reported in wild geese and mallards within the CDARB.

PTA020 Effects of Lead-Contaminated Sediment on Mallard Duckling Behavior. Douglas-Stroebel, E.*, Brewer, G.L., Frostburg State University, Frostburg, MD; Hoffman, D.J., USGS Patuxent Wildlife Research Center, Laurel, MD. Ingestion of lead-contaminated sediment, related to past mining activity in the Coeur d'Alene River Basin (CDARB) in Idaho, was evaluated for effects on behavior of mallard ducklings, using time-activity budgets over a five-week period. Day-old ducklings received untreated control diet, clean sediment (24%) supplemented control diet, or CDARB sediment (3449 ug/g lead) supplemented diets at 12% or 24%. Also, the effect of nutrition was evaluated with a less than optimal diet (two thirds corn and one third standard diet), and the form of lead in the sediment compared with lead acetate. The incidence and duration of ten behaviors was recorded (resting, standing, moving, drinking, dabbling, feeding, pecking, preening, bathing, and swimming). Contaminated sediment significantly affected the proportion of time spent swimming, yet did not significantly affect any of the other recorded behaviors. There were also signs of disruption of balance and mobility observed in the 24% contaminated sediment groups and the lead-acetate group. Nutrient level affected the amount of time spent in water-related behaviors, the growth rate, and the initial time of molt. Although the proportion of time spent in behaviors other than swimming was not markedly affected by ingestion of the contaminated sediment, the observed problems with balance and mobility coupled with decreased time spent swimming illustrate a potential threat of the contaminated sediment to the survival of mallard ducklings in the wild.

PTA021 Heavy Metals Toxicity in the American Dipper (*Cinclus mexicanus*). S.M. Strom, H.S. Ramsdell, Center for Environmental Toxicology and Technology, Department of Environmental Health, Colorado State University, Ft. Collins, CO and A.S. Archuleta, U.S. Fish and Wildlife Service, Lakewood, CO. Historic and ongoing hard rock mining operations near the headwaters of the Arkansas River in Colorado have resulted in elevated concentrations of dissolved and sediment bound metals including lead, copper, cadmium and zinc. The invertebrate community consists of metal tolerant species that have the ability to accumulate metals. Therefore, it is believed that invertebrate predators such as the American dipper, *Cinclus mexicanus*, may be exposed to elevated levels of metals through food chain transfer. Since dippers are dependent on aquatic life, they will make an excellent sentinel species, and be a useful indicator for monitoring contamination and recovery of metal impacted streams. Dippers from moderately impacted and heavily impacted streams had significantly lower blood aminolevulinic acid dehydratase activity (828 and 428 nmol porphobilinogen/ml red blood cells/h respectively) compared to dippers from non-impacted streams (1084 nmol PBG/ml RBC/h). These results indicate that dippers from impacted streams are being affected by elevated lead concentrations in their prey.

PTA022 Effects of Environmental Contaminants on Wildlife Using Resacas and Settling Basins in the Lower Rio Grande Valley, Texas. Wainwright, S.E.*, Texas A&M University, College Station, TX; Mora, M.A., Texas A&M University, College Station, TX; Thomas, P., University of Texas Marine Science Institute, Port Aransas, TX; Sericano, J., Texas A&M University, College Station, TX; Donnelly, K.C.,Texas A&M University, College Station, TX. Ninety -five percent of the Taumalipan brushland ecosystem in the Lower Rio Grande Valley (LRGV) of Texas has been cleared for agricultural, urban, and industrial development. The remaining habitat supports more that 700 vertebrate species, 86 of which are endangered. Many wildlife species rely on small oxbow lakes (resacas) and shallow artificial reservoirs (settling basins) for aquatic habitat. Most of these water bodies receive pesticide drift and urban runoff, as well as treated and untreated sewage inputs. Potentially estrogenic contaminants, such as phthalate esters and chlorinated pesticides, have been identified in water, sediment, and fish at these sites. The main objective of this study was to determine if reproduction in wildlife using LRGV resacas and settling basins is being affected by such contaminants. In the spring and summer of 1997 we followed avian use and reproductive success at 5 resacas and 5 settling basins. Water and sediment from these sites were collected and analyzed for OCs, OPs, and phthalates. Common carp (*Cyprinus carpio*) and eggs of the green heron (*Butorides virescens*) were analyzed for OCs. Steroid hormones and vitellogenin in male common carp plasma were measured, and used in conjunction with gonadal somatic index values as indicators of potential exposure to mixtures of estrogenic compounds. Green heron reproduction was considered normal, despite high levels of some chlorinated compounds in eggs. Vitellogenin was found in the plasma of male carp at some sites, and suggests exposure to estrogenic compounds.

PTA023 Environmental Contaminants in Ocelots of the Lower Rio Grande Valley, Texas, 1986-97. Mora, M.A.*, U.S. Geological Survey, College Station, TX; Laack, L., U.S. Fish and Wildlife Service, Rio Hondo, TX; Lee, C.M., U.S. Fish and Wildlife Service, Corpus Christi, TX; Sericano, J., Texas A&M University, College Station, TX; Gardinali, P., Texas A&M University, College Station, TX; Frank, D., Texas A&M University, College Station, TX. The Lower Rio Grande Valley (LRGV) in Texas is home to several endangered and threatened species. The ocelot (*Felis pardalis*) was classified as endangered in 1982, under the endangered species act of 1973. Habitat loss and fragmentation endanger the recovery of ocelots and potential ocelot habitat includes only about 1.6% of the south Texas area. The range and foraging habitat of ocelots includes agricultural areas that contain pesticides. Bird deformities and a reported high incidence of birth defects in humans in the LRGV in the early 1990s prompted several studies to evaluate the potential effects of contaminants on wildlife of the LRGV. The main objective of this study was to determine if ocelots in the LRGV were affected by environmental contaminants such as PCBs, chlorinated pesticides (OCs), and trace elements. Blood and hair of 25 live-trapped ocelots were collected from 1986-1997 for the analysis of OCs, PCBs, and trace elements. Tissues of road-killed ocelots were also analyzed for OCs and metals. Organochlorine compounds in blood plasma and tissues of road-killed ocelots were low and at levels not likely to affect their reproduction. Mercury and Se were detected in 72% of the samples and ranged from 0.5 to 1.25 µg/g dw and 1.5 to 3.5 µg/g dw, respectively. Overall, Hg and Se concentrations in ocelots were much lower than those observed in the Florida panther (*Felis concolor*) in Florida. Low levels of OCs and trace elements in ocelots and their apparent good productivity (mean=1.5 young per litter), suggest that these contaminants are currently not a threat to the survival and conservation of this endangered species in the LRGV.

PTA024 Wildlife Secondary Toxicity Studies with Warfarin. Poché, R.M., Genesis Laboratories, Inc., 10122 N.E. Frontage Road, Wellington, CO 80549. Warfarin was developed in the 1940s as a rodenticide for both urban and field rodent control. After reports of genetic resistance to warfarin in rats and mice from various parts of the world, the development of more toxic and persistent second generation anticoagulants have virtually replaced warfarin. In the U.S., second generation compounds are not registered by the U.S. EPA for field rodent control. Although warfarin is a safer compound and can be efficacious when used against field rodents, little information is available on the potential for secondary poisoning to non-target wildlife that may consume rodents killed by the bait. Laboratory tests were conducted in which 500 ppm warfarin bait was presented to rodents until death. These were fed to European ferrets and black-billed magpies for seven days. Observations were made twice daily on the ferrets and birds during the exposure and 21-day post-treatment periods. No mortality to non-target wildlife was observed. This study demonstrated that warfarin should be re-considered for use in field situations over the more potent products to control pest species, such as ground squirrels, voles, or other rodents.

PTA025 Effects of Dietary PCB Exposure on Food Intake and Metabolic Rate in White-footed Mice (*Peromyscus leucopus*). Voltura, M.B.*, French, J.B. USGS Patuxent Wildlife Research Center, Laurel MD. Energy budgets have provided physiological ecologists with a vital link between environmental variables and individual performance, and should also prove useful to ecotoxicologists in understanding the effects of sublethal exposure in the field. Exposure to toxic compounds is likely to be metabolically expensive, and may result in a trade-off between energy spent to detoxify and excrete contaminants, and energy allocated to growth or reproduction. To examine this energetic cost, we fed captive white-footed mice (*Peromyscus leucopus*) diets containing PCBs (2:1 Aroclor 1242:1254) at levels of 0, 0.1, 10 and 25 ppm. After 6 weeks on the diets, there were no differences in food intake (g/d), diet digestibility (%) or body mass related to the level of dietary PCBs. This indicated that short term exposure to PCBs did not cause a detectable increase in energy need, as measured by voluntary food intake. We continued to feed mice the PCB-containing diets for one year, and then measured resting metabolic rate (ml O_2 consumed per hour) at 30°C. After one year, all mice had gained mass, but mice on the 25 ppm diet were significantly heavier than mice in the other groups. Mice on the 25 ppm diet also showed a significant increase in metabolic rate ($p<0.01$), even after correcting for the differences in body mass. Oxygen consumption for mice on the 25 ppm diet averaged 40.1 ml O_2/h, an increase of 10% over control mice, suggesting that there is an energetic cost to ingesting contaminants. Current studies are examining metabolic rates of PCB-exposed mice at temperatures below 30°C, and during reproduction, to determine whether the effect of PCBs on metabolism is dependent on energy demand.

PTA026 Mink (*Mustela vison*) and Other Mustelid Species as Important Sentinels of Environmental Health Hazards. Sheffield, S.R.*, Clemson University, Pendleton, SC, Osowski, S., EPA Region VI, Dallas, TX. Among the mammalian families most widely studied as environmental sentinels are members of the Family Mustelidae. Much of this work has been conducted with the mink (*Mustela vison*), and it has been clearly demonstrated that mink are highly sensitive to a number of widely distributed environmental contaminants, including PCBs, dioxins and furans, and various metals and pesticides. Studies with mink began in the mid-1960s in response to a noticeable decline in the reproductive success in ranch mink fed Great Lakes fish. Studies with river otter (*Lutra canadensis*) have also shown this species to be sensitive to a number of environmental contaminants. From 1989-1991, 141 mink were collected from SC, NC, and GA and were analyzed for levels of 17 different contaminants. Currently, an analysis of reproductive organs from these mink has shown a relatively strong correlation between tissue levels of certain environmental contaminants (e.g., Hg, PCBs, DDE, dieldrin) and reproductive organ weights and possible reproductive failure. Not surprisingly, mink have all but disappeared along the coastal plain of the southeastern US. Further investigation is being conducted into the role of contaminants in mink declines in the southeast, including the introduction of radiotagged mink from clean areas into areas where mink have disappeared. Life history characteristics of mustelids (high trophic level, relatively high reproductive rates, demonstrated sensitivity to contaminants, biology well known, trapping/handling/anesthesia procedures established, etc.) make them ideal sentinel species of potential environmental health hazards. The history and current state of our knowledge on aquatic mustelid toxicology and the possible role of environmental contaminants in population declines of aquatic mustelids will be presented.

PTA027 Occurrence of Butyltin Compounds in River Otters Collected from Riverine and Estuarine Areas in Washington and Oregon States. *Kannan, K.*, Senthilkumar, K., Grove, R.A., Henny, C., Giesy, J.P., Michigan State University, East Lansing, MI; Department of Environment Conservation, Ehime University, Japan; U.S. Geological Survey, Corvallis, Oregon. Tributyltin (TBT) and its degradation products, mono- (MBT) and dibutyltin (DBT), were measured in liver tissues of adult male river otters collected from several riverine and coastal estuarine locations in Washington and Oregon states. Butyltin compounds were detected in the liver of all the 40 river otters taken from various rivers and the concentrations ranged between 12 and 2600 ng/g, wet wt. Otters taken from Camano Island, Fort Ward, Bremerton and Eglon in Washington state, contained butyltin concentrations greater than 1000 ng/g, wet wt, in the liver. Concentrations were greater in otters collected from areas where there has been intensive boating activities. DBT or MBT was the predominant compound found in river otters. These results suggest the occurrence of butyltin compounds in rivers, possibly arising from the disposal of wastewater or from boating activities.

PTA028 Dose and Time Dependent Changes in Phosphorylation of Cellular Proteins From Pinniped Adipose Tissue Exposed to 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD). Olsen, H.E.*, Elliott, J., University of California, Santa Cruz CA; Haulena, M., Gullen, F., Marine Mammal Center, Golden Gate National Recreation Area, Sausalito CA; Tjeerdema, R.S., University of California, Santa Cruz CA. Little is known concerning the effects of polychlorinated biphenyls (PCBs), polychlorodibenzo-p-dioxins (PCDDs), and polychlorodibenzofurans (PCDFs) on normal adipose tissue function in pinnipeds, who depend on this tissue for insulation, for energy reserves during periods of fasting, and as a source of energy for maternal investment in the young. The objective of the present study was to evaluate the sensitivity of pinniped adipose tissue to the effects of PCBs and PCDDs relative to the laboratory species guinea pigs, mice, and rats using changes in phosphorylation of cellular adipose proteins as an endpoint for comparison. 2,3,7,8-TCDD was used as a model toxicant and changes in phosphorylation state of cellular proteins from adipose tissue of laboratory species compared with three pinniped species, the northern elephant seal (*Mirounga angustirostris*), the California Sea Lion (*Zalophus californiaus*) and the California Harbor seal (*Phoca vitulina richardsi*). Samples of adipose tissue were dosed with a range of 2,3,7,8-TCDD concentrations, incubated 1/2-4 hours, homogenized and centrifuged, proteins phosphorylated, separated by SDS-PAGE, and detected by autoradiography. Tissue proteins exhibited a complex pattern of phosphorylation, dependent on dose of 2,3,7,8-TCDD and incubation time. Two protein bands of approximately 130-110 kDa and 35-30 kDa containing phosphotyrosine residues were induced in all species of seals. No large species differences were observed for the lowest dose of 2,3,7,8-TCDD eliciting a change in phosphorylation state of adipose tissue proteins.

PTA029 Organochlorine Contaminants and Cytochrome P4501A Expression in Blue Whale (*Balaenoptera musculus*) from the St. Lawrence River, Canada. Koenig, B.G.*, Metcalfe, T.L.*, Metcalfe, C.D. Environmental and Resource Studies, Trent University, Peterborough ON Canada; Stegeman, J.J., Moore, M.J., Miller, C.A. Biology Dept. Woods Hole Oceanographic Institution, Woods Hole MA USA; Sears, R. Mingan Island Cetacean Study, Inc., 285, Green, St-Lambert QC Canada. In biopsy samples of skin and blubber from blue whales in the Gulf of St. Lawrence sampled in 1996, cytochrome P4501A expression in dermal endothelia was detected immunohistochemically using MAb 1-12-3. Results were scored on a scale of 0-15, and showed elevated expression in biopsy samples from off Sept Isles (7.2 ± 1.0, n=8) as compared to the Gulf of St. Lawrence (3.7 ± 3.0, n=6, $p<0.005$) and Iceland (1.8 ± 2.0, n=9, $p<0.001$). These data suggest differences in circulating levels of Ah receptor agonists in blue whales sampled from these locations. Organochlorine contaminants (PCBs and OC pesticides) were elevated in the blubber of blue whales from the Sept Isle site sampled in 1997 (e.g total PCBs; $1,746 \pm 620$ ng/g lipid, n=7) as compared to blue whales from other areas of the Gulf of St. Lawrence (355 ± 182 ng/g lipid, n=20). Elevated responses were measured in the extracts from blubber biopsies of Sept Isle whales using the recombinant yeast screen for endocrine disrupting substances. These studies illustrate the utility of using biopsy samples for biomarker and biomonitoring studies of cetaceans.

PTA030 Organochlorine Contamination in Small Cetaceans Stranded Along Florida Coastal Waters, USA. Takahashi, A.*, Ehime University, Matsuyama, Ehime, Japan; Kannan, K., Michigan State University, East Lansing, MI, USA; Loganathan, B. G., Murray State University, Murray, KY, USA; Odell, D. K., Sea World Inc., Orlando, FL, USA; Tanabe, S., Ehime University, Matsuyama, Ehime, Japan. Concentrations of persistent organochlorines were determined in the liver of small cetaceans (bottlenose dolphin, atlantic spotted dolphin, pygmy sperm whale) stranded along Florida coast during 1989-1994. Mean concentrations of PCBs (110 μg/g wet wt.) were the highest followed by DDTs (9.5 μg/g wet wt.), chlordane compounds (CHLs: 2.3 μg/g wet wt.), *Tris* (4-chlorophenyl) methanol (TCP methanol: 0.20 μg/g wet wt.), *Tris* (4-chlorophenyl) methane (TCP methane: 0.11 μg/g wet wt.), HCB (0.052 μg/g wet wt.), and HCHs (0.024 μg/g wet wt.), respectively. Organochlorine concentrations in spotted dolphin and pygmy sperm whale were one or two orders of magnitude lower than those in bottlenose dolphins. Both spotted dolphin and pygmy sperm whale are off-shore species; therefore, the exposure to organochlorines is expected to be minimal. Specific accumulation pattern of PCB congeners was observed in this animals. Especially some bottlenose dolphins exhibited a comparable proportion of octa- and nonachlorobiphenyl congeners. The major PCB formulations such as Aroclors 1016, 1242, 1254, 1260 contain little higher chlorinated PCB isomers, so these results suggest that some dolphins exposed not only to such common PCB formulations, but also to a highly chlorinated PCB formulations like Aroclor 1268. The estimated 2,3,7,8-TCDD equivalents of coplanar PCBs in these small cetaceans were between 160-17000 ng/g fat wt., which indicated that some dolphins had much higher concentrations than those in the U.S. Atlantic coast and Mediterranean Sea earlier reported.

PTA031 Geographical Representation of Toxic Stress to Birds and Mammals due to Heavy Metal Bioaccumulation. Traas, T.P.*; Luttik, R.; Mensink, H.; National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands (tp.traas@rivm.nl). Bioaccumulation of metals in food webs may lead to unacceptable exposure of birds and mammals. An indicator of toxic stress is the fraction of birds and mammals species, that are exposed to diet concentrations above the No-Effect Diet Concentration. This fraction is called the Potentially Affected Fraction (PAF) and is used as an indicator of toxic stress on birds and mammals. With the PAF, a common denominator is found for aggregating risks of different heavy metals. To calculate toxic stress to birds and mammals, several difficulties must be circumvented: i) The effect of natural background concentrations of heavy metals; only the effect of added metal (of anthropogenic origin) was considered. ii) Bioaccumulation of metals in the food web; bioaccumulation is highly dependent on location- and species specific bio-availability. iii) Laboratory toxicity data must be extrapolated to toxicity to birds and mammals in the field; correction factors for caloric content of food, food assimilation efficiency and metabolic rate were applied. Starting from soil concentrations, concentrations in several diet items were calculated. For selected birds and mammals, the Potentially Effect Concentration in the diet could be compared to the estimated No-Effect Concentration in the diet. If the PEC is larger than the NEC, the species was considered at risk. The fraction of selected species with PEC > NEC is the PAF for birds and mammals. Maps of the PAF were calculated for copper, zinc and cadmium. High PAFs for cadmium can be found on the sandy soils in the Netherlands. The PAFs for copper are negligable. Elevated PAFs for zinc are only found in the western peat areas.

PTA032 The New Avian Dietary Study - Beauty or Beast? Robert Luttik* and Alison J. Johnson, National Institute of Public Health and the Environment, The Netherlands and Huntingdon Life Sciences, England. Approaches to avian toxicity testing were comprehensively reviewed by a SETAC/OECD Workshop at Pensacola in 1994. The workshop report included the first draft of a proposed new guideline for the avian dietary study or LC50 to replace the existing OECD method. Several elements proved controversial: the test period was extended from 5 to 21 days, a change to individual housing required increased accommodation, and a host of unfamiliar sublethal endpoints were proposed. Many feared these changes would greatly increase the cost of the study. But it is important to remember the shortcomings of the existing test. For many chemicals the result is confounded by severely reduced intake of the treated food, so that supposedly different treatments result in the same exposure. In some cases the test is measuring starvation rather than toxicity. The 5-day test period frequently fails to represent the full duration of exposure in the field, for which the true LC50 may be lower. Altogether the existing test lacks ecological relevance and may produce an inappropriate risk assessment. The latest draft of the new guideline overcomes these problems. The duration of the test is flexible, between 5 and 21 days, according to need. The primary endpoint in the proposed dietary study is mortality. When sublethal endpoints are required, they would be better assessed in an avian reproduction study. Individual food consumption will be measured to provide a true measure of exposure and allow the termination of treatment levels likely to cause starvation: a significant welfare benefit. Thus the perceived disadvantages of the Pensacola draft have been removed, and the latest draft offers an ecologically-relevant test which meets the needs of risk assessment.

PTA033 Effects of Cadmium Pollution on *Mus musculus* Populations in Estuarine Systems using Metallothionein as a Biomarker. Tikkanen, L.*, San Diego State University, CA, McClenaghan, L., San Diego State University, CA. NPS pollution is a chronic problem for estuarine environments near municipalities. The study site, Tijuana River National Estuarine Research Reserve, has cadmium containing NPS pollution carried to it via San Diego storm drains and the Tijuana River flowing from Mexico. Little is known of the effects Cd has on resident small mammal populations and whether or not it has progressed even farther through the food web. The objectives of this study were to quantify Cd contamination at Tijuana Estuary in San Diego California, determine Cd's effects in resident *Mus musculus* populations, and evaluate theapplicability of using metallothionein (MT) as a heavy metal biomarkerin *Mus musculus*. We have evaluated Cd levels in plant, soil and Mus musculus tissue samples. Hepatic and renal damage were evaluated based on ALT/AST and BUN protein levels in serum samples. Testes weights were taken for the males and uterine scars enumerated in female Mus. MT levels were evaluated and analyzed for a correlation between tissue Cd levels and elevated MT levels.

PTA034 Probabilistic Calculation of a Risk-based Performance Goal for Wading Birds in the Southeastern United States. Preziosi, D.V.*, Durda, J.L., The Weinberg Group Inc., Washington, DC; McMurray, S., LaPoint, T., TIEHH, Texas Tech University, Lubbock, TX. A probabilistic exposure analysis model was developed within a risk-based framework to determine safe levels of the organochlorine pesticide DDT in the prey of wading birds foraging in southeastern freshwater wetlands. Probabilistic approaches (e.g., Monte Carlo) have not reached widespread acceptance in ecological risk applications, particularly within the regulatory sphere, despite the more plausible estimations of risk they provide. In probabilistic analyses, point estimates in a model equation are replaced with probability frequency distributions which account for variability in input parameters. The novel approach developed as part of this probabilistic model offered the distinct advantage of incorporating the inherent variability in behavior that occurs within natural wading bird populations such that the range of theoretically possible exposures could be evaluated. Three exposure parameters were expressed using independent probabilities based on key behavioral attributes: (1) foraging area, (2) site fidelity, and (3) foraging time. Further, the model was constructed within the hazard quotient (HQ) framework, whereby risk was expressed as the ratio of exposure to toxicity. The output of this model was a cumulative frequency distribution of safe levels of DDT in prey, from which percentile frequencies were tallied, and estimates of dietary levels protective of wading bird populations were derived. As probabilistic ecological risk applications gain increasing acceptance, this model and others like it may be used to more accurately predict exposure and risk in naturally occurring animal populations by accounting for inherent variability in biological attributes.

PTA035 Estimating Soft Tissue Mercury Concentrations and Speciation Using the DMPS Chelator Challenge. Adair, B.M.[1*], Rummel, K.T.[1], Woods, J.S.[2], Cobb, G.P.[1] and M.J.Hooper[1]. [1]TIEHH / Texas Tech University, Lubbock TX. [2]University of Washington, Seattle, WA. Mammals inhabiting contaminated sites can be exposed to low levels of heavy metals, such as mercury. Even with substantial tissue accumulation, little mercury is depurated in the urine, making the use of urine as an indicator of low level exposure impossible. Chelators, such as DMPS, have been used successfully to increase metal depuration, thereby providing a non-lethal estimation of soft tissue burden. Adult female prairie voles (*Microtus ochrogastor*) were divided into six dose groups. Four groups received drinking water with 1 ppm methyl mercury hydroxide and two received distilled/deionized drinking water. All groups were injected IP with saline or 0.01, 0.1 or 1.0 mmol/kg DMPS. Animals were exposed to mercury for four weeks. Urine was then collected for 24 hours before and after chelator administration. Kidney, urine, and brain samples were analyzed for inorganic and organic mercury using their distinct reduction potentials. Vole Renal organic mercury concentrations were 10 fold higher than renal inorganic mercury concentrations. Renal organic, but not inorganic mercury, decreased in a dose dependent manner through increased DMPS administration. Urinary organic mercury was approximately 2 fold higher in concentration than inorganic. DMPS treatment caused a dose dependent increase in the urinary depuration rate of mercury. In the highest DMPS dose group, the percentage of post-chelation mercury that could be accounted for by renal depuration approached 30 percent. Dose dependent relationships between urine and soft tissue concentrations will be discussed. These results were used in combination with porphyrin profile analysis to assess trace exposure of voles to mercury. Funded by NIH ES04696.

PTA036 Porphyrin Profile Alteration and DMPS Chelator Challenge Used in Conjunction as a Non-lethal Indicator of Heavy Metal Exposure. Rummel, K.T.*[1], Adair, B.M.[1], Woods, J.S.[2], Cobb, G.P.[1] and M.J. Hooper[1]. [1]TIEHH / Texas Tech University, Lubbock TX. [2]University of Washington, Seattle, WA. Porphyrin profiles are the relative concentrations of heme precursors found in eukaryotic tissue and excreta. Environmental contaminants have been shown to cause alterations in these regularly consistent concentration patterns. DMPS has been used to increase urinary depuration of heavy metals in laboratory animals and human subjects. In studies conducted with human subjects, and more recently with wildlife rodents in this laboratory, it has been found that the combination of the porphyrin profile technique and DMPS chelation is a more effective, non-lethal method for assessing low level exposure and effects of heavy metals. Adult female prairie voles (*Microtus ochrogastor*) were divided into six dose groups. Four groups received drinking water with 1 ppm methyl mercury hydroxide and two received distilled/deionized drinking water. All groups were injected IP with saline or 0.01, 0.1 or 1.0 mmol/kg DMPS. Animals were exposed to mercury for four weeks. Urine was then collected for 24 hours before and after chelator administration. Porphyrin concentrations were determined in kidney and urine samples for each animal. Mercury residues were determined in kidney, brain and urine collections. Methyl mercury exposure led to an increase in 4-carboxylporphyrin in both the kidney and pre-chelation urine, when compared to controls. Increasing levels of DMPS led to a decreasing trend in renal 4-carboxylporphyrin concentration. The ratio of pre-chelation to post-chelation urinary 4-carboxylporphyrin concentration increased in a dose dependent manner as well. Pre-chelation porphyrin profiles correlated best with post-chelation mercury concentrations. These data support the utility of using both the porphyrin profile and the DMPS chelation techniques, in concert, to assess trace methyl mercury exposure in voles. Funded by NIH ES04696.

PTA037 Estimation of Exposure and Effects from a Mine Tailings Release Upstream of Endangered Species Critical Habitat. Gruber, D.*; Rasnake, W.J.; Rinehardt, M.L.; Wilhelm, E.S.; Yeager, J.L.; Biological Monitoring, Inc., Blacksburg, VA. In 1996, approximately six million gallons of clarified water and mine tailings were accidentally released. The discharge occurred upstream of an area containing several species that are Federally listed as endangered. Furthermore, the area is a designated critical habitat for Federally endangered fish. An initial fish kill, in the upper reaches of the receiving system, was estimated at 11,400 by the State regulatory agency. However, further downstream, where the threatened and endangered species exist, no acute toxic effects were noted. Shortly after the release, efforts were made to remediate the streams and bank areas. Subsequently, an extensive ecological assessment of the system was initiated to characterize the potential for short and long term effects. The emphasis has been placed on characterizing both the exposure and the effects to indigenous organisms. Modeling techniques have been used in an attempt to characterize the exposure and transport of the released materials. Chemical and biological assessments have been used to assess the potential for effects. U.S. EPA ECOTOX values have been applied to assess the chemical data. Biological assessments have utilized both laboratory and field techniques. To date, the data have yet to suggest the need for further remediation.

PTA038 A Possible Relationship Between Macroinvertebrate Community Structure and Tissue-Metals Below Mill Tailings, Soda Butte Creek, Montana. Nimmo, D. R.*, MidContinent Science Center, BRD/USGS, Ft. Collins, CO; Castle, C. J., Colorado State University, Ft. Collins, CO. Tissue-metals and community structure of macroinvertebrates above and below mill tailings in Soda Butte Creek, Montana were analyzed in the fall of 1995 and 1996. Data from nine sites in the Creek influenced by the tailings were compared to each other and to data from a reference stream not influenced by mining. Using tissue-metal concentrations compared to EPT and the numbers of species as indicators of community *health*, 52 mg/kg of copper (dry weight) and 172 mg/kg of zinc (dry weight) appeared to be detrimental to the macroinvertebrates. Surprisingly, concentrations of tissue-metals varied significantly from one year to the next and the reasons for the variation are discussed.

PTA039 A Toxicological Reconnaissance of the Upper Animas River Watershed, Silverton, CO. Nimmo, D. R.*, MidContinent Ecological Science Center, BRD/USGS, Ft. Collins, CO; Castle, C. J., Colorado State University, Ft. Collins, CO. Toxicity tests were conducted on surface and subsurface (interstitial) water from the main stem and tributaries of the Animas River. Test species were rainbow trout, *Oncorhynchus mykiss*; fathead minnows, *Pimephales promelas*; daphnids, *Ceriodaphnia dubia*; and amphipods, *Hyalella azteca*. Initial tests were conducted with undiluted waters then detailed tests were conducted on dilutions of the most toxic sites. Subsurface water from A72, a point below the confluence of all the tributaries was the most toxic with the LC50s ranging from the lowest of 31% for the daphnids followed by 35% for the fathead minnows, 40% for the trout and 56% for the amphipods. Covariated analysis of the data for fish suggests that both copper and zinc are responsible for the toxicity at A72 whereas multiple samples of surface water from Elk Park, showed intermittent toxicity to daphnids due to fluctuating copper.

PTA040 Heavy Metals in the Rio Negro: A Comparison Analysis of Flood and Low Water Levels. Paulos, P.M.* and C.L. Howard, University of Houston – Clear Lake, Houston, TX, USA. The Rio Negro, the largest tributary of the Amazon River, serves as the principal route of transportation to the most populated area of Amazonia. This region continues to experience an influx of human habitation, with an increasing degree of agriculture and small-scale industrialization introducing a variety of pollutants to the riverine system. The research as presented endeavors to 1) provide information concerning the presence of heavy metals in the Amazon Basin and 2) provide a comparative analysis between high water and low water levels in the river. Fifteen sites were sampled, with water being collected at surface and at depth. Emphasis was placed on sampling in the vicinity of the industrialized port city of Manaus, Amazonas, Brazil, which is situated at the confluence of the Rio Negro and the Amazon rivers. Additionally, sediment samples were obtained, where feasible, by use of a grab sampler. Upon collection, all samples were preserved according to prescribed EPA methods and transported to the United States for analysis. The samples were analyzed using Inductively Coupled Argon Plasma (ICP) methods for lead, cadmium, copper, chromium, nickel, and zinc. These metals were chosen for analysis because of their proven adverse effects on the aquatic environment when present at elevated levels. Additionally, iron, calcium, magnesium, and aluminum were analyzed via ICP. For the metals analyzed, the ICP was reproducible at 0.005 to 0.01 mg/L, a level deemed acceptable due to the nature of this study. The study was initiated with the expectation of identifying various heavy metals at elevated concentrations and recognizing a relationship between low water and flood stage metal concentrations. Obtaining data on heavy metal levels in the Amazon Basin provides a base on which future research can be conducted. This will provide a greater understanding of the burden that is being placed upon this vast, fragile system.

PTA041 Use of Macroinvertebrates for Biocriteria Evaluation of Eagle River: Selection of Indices. J.S. Volosin* and R. D. Cardwell, Parametrix, Inc., Kirkland, WA. An evaluation of the effectiveness of cleanup efforts to restore biological health of the Eagle River, Colorado is being done to address zinc mining-related impacts. Indices for the evaluation of macroinvertebrate health are being selected for use as biocriteria. Seventeen indices were evaluated using seasonal data from 1993 to 1997. The appropriateness of indices was evaluated using correlations and box plots. When two indices were highly correlated, one of the indices was eliminated from further analyses. The remaining indices were evaluated using box plots. The overlap of box plots between reference and monitoring sites for each index was evaluated and scored. The more box plots overlapped for an index, the higher the score. Spring data resulted in little overlap between reference and monitoring sites for total taxa richness, number of mayfly taxa, percent mayfly, and percent collector gatherers. This indicates differences between locations for these indices in the spring. These observations were not apparent with the summer and fall data. Based on the difference observed for the spring data, it was concluded that the above indices are appropriate for evaluating the Eagle River data, and will allow objective evaluation of the effectiveness of Eagle Mine remedial efforts.

PTA042 Trifluoroacetic and other Haloacetic acids (HAAs) in Snow Samples with Emphasis on an Altitudinal Transect from the Rocky Mountains. Martin, J., University of Guelph, Guelph ON; Muir, D.C.G., Scott, B.F., Spencer, C., Environment Canada, Burlington, ON; MacTavish, D., Atmospheric Environment, Downsview, ON;. von Sydow, L. Linkoping University, Sweden; Mabury, S., University of Toronto, Toronto ON; Blais, J., University of Alberta, Edmonton AB. Trifluoroacetic acid (TFA), is a very stable degradation product of HFC and HCFC replacements for ozone-destroying chloro-fluorocarbons. TFA concentrations in the atmosphere have been predicted to increase 10x as a result of the growth in use of its HFC/HCFC precursors for auto air-conditioning and refrigerant uses. HAAs, especially monochloroacetic acid (MCA)- and trichloroacetic acid (TCA) have been identified as an environmental concern in Europe over the due to their phytotoxicity. HAAs have low Henry's law constants and are readily scavenged by precipitation. We have determined TFA and other HAAs in snow samples collected from selected areas of Canada. HAAs were determined by GC-MS analysis of the anilide derivatives. A series of samples were collected on a transect of increasing elevation (700 m to 2400 m) in the Canadian Rocky Mountains near Banff (AB). These samples contained TFA (25 ng/L), MCA and TCA, with the dominant component being dichloroacetic acid (150 ng/L). There was a general trend of increasing concentration as the elevation increased. HAA levels were generally higher in samples of snow from Ontario. Snow samples from the Turkey Lakes area, east of Lake Superior, contained TFA at concentrations of 350 ng/L while snow from the Lake Simcoe area had considerably lower values (10 ng/L). The results suggest that regional variations HAAs may be related to distance from urban sources, altitude and mean air temperature.

PTA043 Spatial and temporal variability in whole-organism characteristics of reference populations of white sucker (*Catostomus commersoni*) used for cumulative effects monitoring. Gibbons, W.N.*, Munkittrick, K.R., McMaster, M.E., Environment Canada, Burlington, ON, Canada, Portt, C.B., C. Portt and Associates, Guelph, ON, Canada, Van Der Kraak, G.J., University of Guelph, Guelph, ON, Canada. White sucker has been used as a sentinel species for monitoring cumulative effects within the Moose River Basin in northern Ontario, Canada. The basin is the site of several developments including pulp and paper mills, peaking and run-of-the-river hydroelectric stations, mining operations and municipal waste water facilities. In an effort to conduct cumulative effects monitoring it was necessary to document the baseline or reference status of white sucker populations prior to evaluating whether exposed populations are influenced by the development. Several comparisons were conducted to assess reference fish characteristics: 1) longitudinal comparison evaluated the downstream variability in whole-organism characteristics of white sucker within a reference river, 2) a multi-river comparison was conducted to assess differences in reference populations from different river systems within the basin (of the same latitude); and, 3) temporal comparisons were made to document natural changes in characteristics of sucker over time. Results from this work highlighted the degree of variability in whole-organism characteristics of white sucker populations on a spatial and temporal scale. The information was particularly important when determining whether fish populations downstream of industrial and municipal discharges were being affected relative to reference populations, and to decide, on a basin-wide scale, whether additional development within the basin would be acceptable.

PTA044 Monitoring Water Quality in Urban Environments. Forsythe II, B.L.*, Belden, J., Burdette, J., Carrier, A., Ternes, M.J., Lydy, M.J., Wichita State University, Wichita, Kansas. Nonpoint source pollution of surface waters has come under increasing scrutiny. Urban areas with their wide range of land-use activities provide numerous sources of pollution that can be categorized as nonpoint in nature. This study was designed to assess the water quality associated with four major types of land-use activities: agriculture, golf courses, parks/recreational areas, and general urban sites. Common water quality parameters and concentrations of select pesticides were measured on a monthly basis at various sites in Wichita, KS. Generally the water quality at each site was adequate to support aquatic life. One site with significant groundwater input was found to have consistently low concentrations (< 0.5 μg/L) of triazine herbicide(s). The sites receiving runoff from streets and general urban areas were found to have the most impaired water quality. Concentrations of nitrate and total phosphorus peaked in the fall and spring, coinciding with increased fertilizer applications on lawns and golf courses. Changes in water quality were directly related to the amount of precipitation associated with rain events and the time between events.

PTA045 The Use of Automatic Run-off Sampling Equipment. Bright, S.A.*, Berger, R. G., Green, M.T. USDA-APHIS, Riverdale, MD; W.G. Eastland, Prince George, BC, Canada. Effective environmental monitoring of USDA programs that use pesticides requires that overland run-off from rainstorms be sampled for residues. Sampling is logistically difficult, thus, USDA personnel developed the automatic run-off sampling equipment (A.R.S.E.), allowing for passive run-off water collection. An A.R.S.E comprises a 500 ml vessel placed into the ground, within a run-off channel. A screen over the vessel excludes pebbles or dirt clods. A cover raised 4 above ground level prevents rain or applied pesticide from directly entering the vessel. Five years of A.R.S.E. data, collected as part of the National Boll Weevil Cooperative Control Program, yielded malathion residues that averaged 14.1 ppb (n=113, range 0.1-185 ppb, median = 2.5). Run-off samples were collected near the treated field edge. Additional samples collected at the edge of water bodies near treated fields averaged 5.4 ppb (n=49, range 0.1 ppb - 93.4 ppb, median = 0.5). Field edge samples had significantly more malathion than water edge samples (p = 0.016). The data showed a significant correlation between the amount in a field edge sample and the corresponding sample collected at the water edge (n = 98, corr. coef. = 0.848). In addition, the amount of malathion in run-off collected at the field edge was significantly greater when the rainfall occurred within one day of treatment, as compared to when rainfall occurred more than one day after treatment (p = 0.002). However, elapsed time between treatment and rainfall was not a reliable predictor as to how much residue would be found in run-off water at the field edge ($r^2 = 0.063$).

PTA046 Assessment of Water Quality in Wichita, Kansas Using Fish Community Data Linked with Sediment and Fish Tissue Organochlorine Analysis. Eaton, H.J.* and Lydy, M.J., Wichita State University, Wichita, KS. The Arkansas River and its tributaries in Wichita, Kansas have historically been contaminated with high levels of organochlorine insecticides (OCs), namely chlordane, due to non-point source pollution. Because of this contamination, fish consumption advisories have been effect (1986-1988, and reinstated in 1993) for many areas of Kansas, including within the city limits of Wichita. The surface water areas included in this study represent habitat for endangered and threatened wildlife, as well as for recreational fishing. Water quality assessments included analysis of OC levels in both sediment and fish tissue samples using GC/ECD in addition to an assessment of the fish community structure and overall health by implementation of an Index of Biotic Integrity (IBI). Fish tissue analyses were conducted on species from three trophic levels, the common carp (*Cyprinus carpio*), gizzard shad (*Dorosoma cepedianum*), and bass (*Micropterus* sp.), in order to evaluate possible biomagnification processes.

PTA047 Chena River Aquatic Assessment, Fort Wainwright, Alaska. Burgess, R.M., Jacobs, L.L., ABR, Inc., Fairbanks, AK; Ohlendorf, H.M.*, CH2M HILL, Sacramento, CA; Oswood, M.W., University of Alaska, Fairbanks, AK; Wallace, M.N., Deardorff, T., and Fosbrook, C., U.S. Army, Anchorage, AK. The Chena River Aquatic Assessment was conducted during 1997 (and is being continued in 1998) in support of the Postwide Risk Assessment for Fort Wainwright, AK. The primary tasks of the aquatic assessment were to collect and analyze sediment and benthic macroinvertebrate samples from the Chena River with the goal of assessing impacts of contaminant releases from Fort Wainwright. This assessment was conducted because contaminant concentrations in sediment, especially those related to fuel seepage to the river, exceeded screening-level ecological benchmarks. Kick-net and artificial substrate samples from the segment of the river potentially affected by fuel seepage were compared to samples from similar habitats at upstream reference locations. Using a broad array of Rapid Bioassessment Protocol metrics (primarily with identification to the family level), the methods statistically differentiated among sampling locations. However, macroinvertebrate communities were strongly dominated by Chironomidae, and it was not clear that differences could be directly attributed to contaminants. More detailed identification of the chironomid specimens is planned, because they are likely to provide better characterization of differences among the sampling locations. Organochlorine pesticides and petroleum-related semivolatile organics were detected only in samples from the fuel-seepage portion of the river.

PTA048 Biological Diversity at the UK GREAT-ER Sampling Sites. Webb, S.F.*, Procter & Gamble Technical Centres Ltd., Staines, UK; Williamson, S., Manchester University, UK. Preliminary attempts have been made to discern the potential impact of key physico-chemical/habitat parameters (including LAS) upon the biological diversity at the UK GREAT-ER sites in Yorkshire (UK). Measurement of biological diversity at sampling sites (n = 21) on the rivers Aire and Calder was undertaken in June/July 1997. Observed variations in BMWP (Biological Monitoring Working Party) and ASPT (Average Score Per Taxa) biotic indices were then compared with antecedent measured water quality data (1996-1997) and habitat parameters. This entailed the use of multivariate statistics. The first step involved a factor analysis to group the various physico-chemical/habitat parameters into factors to overcome any inter-correlation. A five factor solution described 93% of the variation in the original data set. The factors themselves generally appear to reflect established/logical relationships (e.g., the interdependency of distance from source, flow, depth and width). These factors were then regressed against BMWP and ASPT scores. R^2 values were 65% and 81% respectively. Factor 5 containing suspended solids was the best single determinand (assuming causality) of BMWP score. For BMWP, the next most important factors were Factor 1 (BOD, Ammonia, Nitrite, LAS & Ortho-Phosphate) and Factor 3 (Distance from source, Flow, Depth & Width). For ASPT, Factor 2 (Chloride, Magnesium, Conductivity, TON & Altitude [-]) was ranked highest in the hierarchy. Factors 3, 5 and 1 were ranked next. Overall, the results show a weak correlation between LAS (via Factor 1) and biological diversity which cannot be separated from the dependence of biodiversity upon the general organic loading. LAS *per se* was not significantly correlated with either BMWP or ASPT. Mean LAS levels at the sites varied from <1 - 32 µg/l.

PTA049 Interlaboratory Performance Analysis In the Determination of Total Selenium in Water: Implications for Water Quality Criteria. Steinhoff P.J., Smith, B.W., Warner, D.W. and Möller, G., University of Idaho, Moscow, ID. This study explores the performance of experienced laboratories for analysis of total selenium in water by a variety of analytical methods. The goal of the study is to examine intra- and inter-laboratory data variability. Replicates (n = 7) of 7 sample types that included known Se concentration references, natural waters and treated waste waters were submitted to seven laboratories with pre-qualified selenium analytical experience. Results of the study indicate large ranges in minimum and maximum results, distinct differences in laboratory precision, and routine reporting of numerical results below statistical limits of quantitation. Hydride generation as a sample introduction technique, demonstrated superior performance. In general, the study supports a caution advisory of using low-level selenium data, especially results less than about 10 µg-Se/L, without quantifying the statistical uncertainty of the data. Since this study used data from samples that were submitted in bulk to participating laboratories and those laboratories were pre-qualified for selenium analytical expertise and experience, the study can be considered a best case demonstration of performance. The regulatory interpretation of data submitted in compliance with the current 5 µg-Se/L chronic water quality criteria should be guarded in the absence of statistical validation of the raw data.

PTA050 The Environmental Fate of Chlorpyrifos and Chlorothalonil in Cranberry Bog Environments. Putnam, R.A.* , Clark, J.M., University of Massachusetts, Amherst, MA. Chlorpyrifos (CHP), equivalent to 2.4 L/ha and two applications of chlorothalonil (CHT), each equivalent to 6.5 L/ha Bravo 720® were applied in the presence and absence of a spreader-sticker adjuvant to plots of "Howes" cranberries in established cranberry bogs. Residues levels of both parent compounds and several environmental metabolites were assessed on harvested fruit and fruit collected over the growing season, on foliage samples collected at 2 h, and 3, 7, 15, and 30 days post application, and in soil samples collected at harvest. Residues from extracted field samples were analyzed by GC/MSD, GC/ELCD, and GC/NPD. Polar metabolites were chemically derivatized with ethyl iodide or diazomethane prior to GC analysis. The majority of residues remaining in cranberry fruit at harvest (76 days post-application) were as CHT (i.e., 46 - 79 ppb), with levels of the CHT metabolites 4-hydroxy-2,5,6-trichloroisophthalonitrile and 1,3-dicarbamoyl-2,4,5,6-tetrachlorobenzene (III) accounting for 36 % and 6 % of the total residues, respectively. The majority of CHT residues in the soil 76 days post-application were as compound III (i.e., 314 - 389 ppb). Smaller quantities of several other CHT metabolites were also detected. Only chlorpyrifos (339 - 422 ppb) was detected in the fruit at harvest (62 days post-application), but residues of chlorpyrifos-oxon and 3,5,6-trichloro-2-pyridinol were detected in soil and foliage. Several of the environmental metabolites products identified in the field studies were also detected in the pesticide formulations Bravo 720® and Lorsban 4E®. The dissipation of dislodgeable foliar residues of CHP and CHT followed first-order kinetics ($r^2 > 0.872$), with estimated half-lives of 3.5 d and 12.7 d, respectively. The addition of the spreader-sticker adjuvant increased fruit and foliar residues, but it did not alter dissipation rate or metabolism.

PTA051 A Preliminary Study of Chlordane in Fish from Connecticut Lakes. Perkins, C.R.*, Neumann, R.M., Carley, R.J., Anderson, M., and Chirdon, S. University of Connecticut, Storrs, CT. Chlordane, unlike many contaminants, is ubiquitous in the environment, resulting in its detection in rainwater, drinking water, air, surface waters, soils, sediments, fish, and humans. There is, at present, a lack of information on residues of chlordane, its isomers, and degradation products in fish from Connecticut lakes and ponds. A limited survey of five lakes was undertaken in Connecticut and fish sampled from Brewster Pond in Stratford had concentrations high enough (relative to the FDA action level of 0.3 ppm) to warrant the issuing of a fish consumption advisory. Brewster Pond is surrounded by residential land use and contamination is likely a result of residential pesticide use. The objectives of this study are: 1) To undertake preliminary monitoring of total chlordane levels of predatory fish from Connecticut water bodies; 2) To determine differences in chlordane concentrations in predatory fish among Connecticut lakes and ponds with varying land use patterns (residential vs. forested). To accomplish these objectives, 9 large largemouth bass (or other suitable species) will be collected at fifteen lakes in Connecticut of two distinct land-use patterns (12 residential and 3 forested/non-residential).

PTA052 Herbicide Contamination in Streams of a Claypan Soil Watershed: Evaluation of the Semipermeable Membrane Device as an Integrative Monitor. Orazio, C. E.*[1], Lerch, R. N.[2], Blanchard, P. E.[3], Petty, J. D.[1], Gale, R.W.[1], Huckins, J. N.[1], Lebo, J. A.[1], Alvarez, D. A.[3]. USGS Columbia, MO [1]; USDA-ARS, Columbia, MO [2], University of Missouri, Columbia, MO[3]. The herbicides atrazine, alachlor, cyanazine and metolachlor are common surface water contaminants in Missouri resulting from corn acreage runoff, especially in claypan regions. USGS and USDA-ARS have on-going programs to monitor herbicides in Missouri streams by taking periodic water samples. Intermittent runoff events can introduce error into the herbicide runoff assessment if high concentrations associated with the events are not measured. We evaluated the semipermeable membrane device (SPMD) as an medium for monitoring herbicides in the streams that are routinely monitored by USGS and USDA-ARS. Grab water samples were taken at the beginning and end of a two week period during which SPMDs were deployed. The SPMDs were of the common design: a thin layer of lipid (triolein) enclosed in a layflat polyethylene tube. The four herbicides accumulated in the SPMD sampler at amounts consistent with factors controlling SPMD uptake and dissipation rates. Due to the herbicides' relatively high water solubilities, and corresponding low K_{ow}s, their accumulation by the SPMD is much less than for compounds such as chlordane, DDT and other higher K_{ow} chemicals. Detailed results of this study will be presented.

PTA053 Nutrients and their Impacts on the Canadian Environment - A National Assessment. Gagnon, C.*, Kent, R.A., Roberts, E.S., Guidelines & Standards Division, Environment Canada, Ottawa, ON, Canada; Chambers, P.A., National Water Research Institute, Environment Canada, Saskatoon, SK, Canada; Charlton, M.N., National Water Research Institute, Environment Canada, Burlington, ON, Canada. The issue of excess nutrients entering the environment from human activities has long been recognized as a significant contributor to environmental degradation, particularly in aquatic ecosystems. Nitrogen and phosphorus nutrient-related impacts (e.g., eutrophication) from agricultural practices, wastewater and industrial effluents are well established in Canada and throughout the world. To date, the sole federal regulatory response under the Canadian Environmental Protection Act relate to a control of phosphorus in laundry detergents. As a result, the Government of Canada has recently commissioned a national assessment to identify the presence and extent of environmental impacts (aquatic and terrestrial) caused by excess nutrients in Canada. The first of its kind, the assessment will attempt to determine whether nutrients as a class, and whether certain nutrients, or nutrient-containing products are problematic and whether those effects are limited to one component of the environment, such as water, or the entire ecosystem, including wildlife. A review of existing regulatory mechanisms for nutrients across Canada is also underway.

PTA054 Environmental effects of application of composted sewage sludge in agricultural production. Győri, Z.*, Kátai, J., Prokisch, J. Debrecen Agricultural University, Debrecen, Hungary. The best way to perform environmental risk assessment of sewage sludge application in the agricultural production is through evaluating long term experiments. Adequate risk assessments can, however, only be performed by considering national and local environment considering e.g. local soils and sludge quality. To perform adequate risk assessments of both the short and long term effects of different sludge management practices, it is necessary to evaluate both chemical and biological impact of the sludge applied soil. A field plot experiments was started at Debrecen Agricultural University in 1997, for studying the long term effect of composted sewage sludge. Evaluating the effects of sewage sludge on soil demands methodology for both chemical and microbiological characterisation. In the chemical test different extraction methods were applied for estimation of amount of metals (Cd, Cr, Ni, Pb, Hg, Zn, Cu, etc.) in different form in soil and sludge. The chemical changes in sludge amended soil are more pronounced in the soil solution than in the soil. Speciation of the soil solution will be a valuable product of the chemical characterisation. Measurement of enzymatic activities in sludge amended will give an overall impression of the bacterial activity. Dehydrogenase is an intracellular enzyme that often indicates the general microbiological activity; while phosphatase and urease also are extracellular enzymes being important for evaluating the mineralisation of phosphorous and nitrogen in the soil.

PTA055 Environmental contaminants in bottom sediments from the Barents and the White Sea. Dahle, S., Akvaplan-niva, Tromso, Norway; Savinova, T.N.*, Akvaplan-niva, Tromso, Norway; Savinov, V.M., Murmansk Marine Biological Institute Russian Academy of Sciences, Murmansk, Russia; Matishov, G.G., Murmansk Marine Biological Institute Russian Academy of Sciences, Murmansk, Russia; Killie, B., Akvaplan-niva, Tromso, Norway. Chlorinated hydrocarbons (α-, β- and γ-HCH, DDT and metabolites, chlordanes, HCB, PCBs 28, 52, 101, 118, 138, 153 and 180, 23 PAH compounds and TBT) have been analyzed in 10 grab samples from Kola Bay (located on the northwest coast of the Kola Peninsula) and Pechenga Bay (the southwest part of the Barents Sea), and in 11 grab and one core samples from the different parts of the White Sea. The amount of sedimentary material less than 63 μm, total organic carbon, total nitrogen were determined in all samples. Chlorinated hydrocarbons and PAHs were determined according Oslo and Paris Commission recommendations (JAMP, 1997), TBT as described by Gremm & Frimmel (1992). Principal component analyses were used to assess contaminant composition differences between stations. Moderate to very high concentrations of TBT, some chlorinated hydrocarbons and PAHs were found in surface sediments from Kola Bay: DDTs levels varied from 3.3 to 10 ng/g d.w., the average levels (6.1 ng/g) was 16 times higher than in the southeast part of the Barents Sea (Loring *et al.*, 1995); PCBs varied from 12.3 to 282.6 ng/g, while in the southeast part of the sea PCB levels do not exceed 0.9 ng/g (Loring *et al.*, 1995). TBT levels varied from 5 to 51 ng/g and were comparable to those found in the Norwegian harbours (Konieczny, 1995). Contamination levels in bottom sediments from Pechenga Bay were moderate, the lowest levels were found in the White Sea, these levels are considered as ecologically safe for benthic animals.

PTA056 Physicochemical, Toxicological and Benthic Monitoring at Dredged Sediments Disposal Site CM-7, Cap-aux-Meules, Quebec, Canada. St-Laurent, D.*, Hamel, P., Provencher, M., Environment Canada, Montreal, QC. Disposal site CM-7 has received through years the non-contaminated dredged sediments of Cap-aux-Meules harbour. These sediments, however, were originally located in harbour areas neighbouring zones with elevated levels of PCBs and PAHs. The main objective of this 1996 environmental monitoring study was to verify if past sediment disposal activities at CM-7 have caused adverse effects to the environment. A bathymetric survey chart was used to position 51 sampling stations at and around disposal site CM-7. Stations were located in the three deposit mounds, as well as in the intermound, near-field and far-field zones. Another 8 stations were placed inside a reference site located approximately 3.5 km from CM-7. All 59 stations were analysed for grain size, metals (As, Cd, Cr, Cu, Hg, Ni, Pb and Zn), TOC, PCBs and PAHs. Twenty-three of these stations were analysed for NH_3-N, redox potential, total sulphides, toxicity (via 5 bioassays), *in situ* bacterial exoenzymatic activity and surveyed for the benthic invertebrate community. No PCBs or PAHs were detected in the samples. Toxicity was observed in the liquid phase Microtox and Sea Urchin assays. However, this toxicity was strongly linked to TOC and NH_3-N, suggesting it was not attributable to anthropogenic contaminants. No toxicity was found in the solid phase Microtox and Amphipod biotests. Laboratory exoenzymatic activity bioassays revealed no toxicity as well but *in situ* exoenzymatic activity was able to distinguish the deposits mounds from the other areas. The benthic community in the deposit mounds was found to have a significantly different structure from that of the other areas, but as productive and healthy. These results and their interpretation will be discussed.

PTA057 Distribution of Sediment Organochlorines in Bays and Estuaries of California. Roberts, C.*, Fairey, R., Jacobi, M. Moss Landing Marine Labs. Moss Landing, CA. Chlorinated pesticides and PCB congeners were measured as part of the Bay Protection and Toxic Cleanup Program's monitoring of sediments in California bays and estuaries. Over 600 samples were analyzed for 36 chlorinated pesticides and 49 PCB congeners at 437 stations throughout California from 1992-1997. These organochlorines were ranked within the data set to compare at the 90^{th} and 95^{th} percentile as well as compared to sediment quality guideline values when available. Major chemical exceedances will be discussed. Two chemical groups having ERM guideline exceedances were total Chlordane and total PCBs. Total Chlordane was measured in 612 samples and exceeded the ERM guideline value of 6 μg/kg in 31% of the samples with the highest concentration being 246 μg/kg. Total PCBs were measured in 684 samples and exceeded the ERM value of 180 μg/kg in 24% of the samples with the highest concentration being19,901 μg/kg. Total DDT was measured in 621 samples and exceeded the ERM value of 46.1 μg/kg in 36% of the samples. Total DDT, normalized to organic carbon, was >100 μg/g OC in only 2% of the samples. Dieldrin was measured in 618 samples and exceeded the ERM guideline value of 8 μg/kg in 5% of the samples with the highest value being 62.6 μg/kg. Spatial patterns and temporal variability in the data will be discussed.

PTA058 PCBs in Fish from Two Maine Atlantic Salmon Rivers. Mierzykowski, S.E.*, U.S. Fish and Wildlife Service, Old Town, ME; Horton, G.E., Maine Atlantic Salmon Authority, Cherryfield, ME; Hathaway, E.M., U.S. EPA, Boston, MA; Carr, K.C., U.S. FWS, Concord, NH. An environmental evaluation of habitat conditions is a critical component in any species recovery program. Two *Downeast* Maine rivers, the East Machias River and Dennys River, support Atlantic salmon *Salmo salar* - a species recently considered for inclusion on the Federal Endangered and Threatened Species List. To assess potential contaminant threats to salmon parr in the two rivers, we collected and analyzed resident fish (*Micropterus dolomieu, Salvelinus fontinalis, Catostomus commersoni*) for PCBs and other environmental contaminants. Forty fish were analyzed for over 90 PCB congeners including non-*ortho* and mono-*ortho* substituted congeners. Total PCB concentrations were highest in wholebody smallmouth bass (0.131 μg/g, WW) and white sucker (0.046 μg/g) from the Dennys River, followed by bass from the East Machias River

(0.023 µg/g). Lower PCB concentrations were detected in brook trout from the Dennys River and white sucker from the East Machias River. The predominant non-*ortho* PCB congeners in bass were 126 and 77, with the highest 126 concentration occurring in Dennys River bass (88.5 pg/g). PCBs in Dennys River bass and suckers were comprised primarily of Cl_5 through Cl_8 substituted congeners. PCB homolog patterns in East Machias River fish were similar to the Dennys River, except Cl_4 substituted congeners were detected in greater amounts particularly in white suckers.

PTA059 Organochlorine pesticide residues in caneland soils - implications for contamination of the near-shore marine environment. Cavanagh, J.E.*, James Cook University; Burns, K.A, Brunskill, G.J. Australian Institute of Marine Science; Coventry, R.J., James Cook University. Organochlorine pesticides were widely used in the Australian cane industry from the early 1950's until 1987 when they were generally banned. The cane industry is located along the coastal margin of northern NSW to far north Queensland. Two river catchments located in North Queensland which contain large areas of caneland are the Herbert and Burdekin river catchments. Erosion of caneland soils and subsequent transport of sediment bound contaminants to the marine environment, especially the Great Barrier Reef, is a major concern. This study has investigated the distribution of organochlorine pesticide residues in near-shore surface marine sediments and surface soils samples and is focussed on understanding the factors controlling the off-site transport of organochlorines. Bulk surface soil samples collected from cane fields in each catchment revealed the presence of easily detectable levels of a-, b, g- and d-HCH, chlordanes, heptachlor, heptachlor epoxide, DDTs, aldrin and dieldrin. Levels found were in the range 0-110ng/g dry wt. Observed differences in the distribution of the organochlorines in soil from the two catchments reflected known historical pesticide usage. Analyses of near-shore surface marine sediments revealed the absence of detectable levels of organochlorines. Initial calculations using known historical application of pesticides (on a catchment basis), estimates of soil erosion from cane-fields and estimates of the total sediment loads of the two rivers suggested that undetectable levels of organochlorines in the marine sediment could be occurring through a simple sediment dilution process. Further work on the distribution of organochlorines in different size fractions of the soil, particle size analyses of sediment in run-off from cane-fields and factors controlling soil erosion processes is currently being undertaken to provide a more accurate model of the off-site fate of organochlorines in a tropical environment.

PTA060 Contamination by Organochlorine Pesticides and Polychlorinated Biphenyls in Sea Otters (*Enhydra lutris nereis*) Found Stranded Along Coastal California, USA. Nakata, H*, Ehime University, Matsuyama, Ehime, Japan; Kannan, K., Michigan State University, East Lansing, MI, USA, Li, J., Ehime University, Matsuyama, Ehime, Japan; Thomas, N., National Wildlife Health Center, Madison, WI, USA; Tanabe, S., Ehime University, Matsuyama, Ehime, Japan; Giesy, J. P., Michigan State University, East Lansing, MI, USA. Concentration of PCBs, DDTs, HCHs, chlordanes (CHLs) and HCB were measured in the liver, kidney and brain tissues of adult sea otters found stranded along coastal California, USA. Contamination pattern of OCs in sea otter from several locations was in the order of DDTs > PCBs >> CHLs > HCHs >> HCB. Hepatic concentrations of PCBs and DDTs were in the range of 58-8700 and 280-5900 ng/g wet wt., respectively, which varied depending on the sampling location. Sea otters collected from Montery harbour contained the greatest concentrations of PCBs, implying local presence of the contamination sources. Among various tissues analyzed, kidney contained higher concentrations. Concentrations of OCs in brain were less, despite its greater lipid content. A noticeable proportion of α-HCH was found in the brain. These could be explained by the specific composition of lipids, occupying higher percentage of polar lipids such as phosphoglycerides and cholesterol in brain. The sex difference in organochlorine concentrations were small, which might be due to less transfer of these compounds to pups through lactation. Sea otter that died from infectious diseases, neoplasia and emaciation contained higher concentrations of PCBs and DDTs than those that died from trauma.

PTA061 The Influence of Life Stage and Cultivar on Basipetal and Acropetal Translocation of Foliar Cd in *Triticum turgidum* L. var durum. Hale, B.A.* and Chan, D.Y., University of Guelph, Guelph, ON, Canada. Durum wheat cultivars can differ quite widely in their accumulation of Cd in grain tissues and their instantaneous rates of root uptake, despite having relatively similar steady-state concentrations of Cd in root tissues. While some of the cultivar differences could likely be explained in terms of acropetal translocation patterns, it is also possible that basipetal translocation could return foliar Cd to the roots, and that this process would be cultivar specific. To examine this hypothesis, that basipetal and acropetal translocation of Cd from durum wheat foliage is independent of life stage and cultivar, plants (*Triticum turgidum* L. var durum cvs Kyle and Arcola) were grown hydroponically in the absence of Cd, and then the foliage was sprayed with cadmium chloride at either 0, 500 or 5000 µg/L at tillering (all foliage), in boot or flowering (bottom half of foliage). Foliage was sprayed each day for three consecutive days; 7.5 mL of solution (including Tween 80, a surfactant) were applied each day. The plants were harvested on the fifth day following the initial Cd spray; the foliage was washed in a solution of HEDTA and Tween 80 prior to harvest. Following acid digestion by the closed teflon vessel technique, tissue Cd concentrations were determined by GF-AAS. The results demonstrated that the cultivars differed in their translocation patterns, in that Arcola, the low grain-Cd accumulator translocated Cd from leaves to roots to a greater extent than did Kyle, the higher grain-Cd accumulator. These patterns were similar across life stages, suggesting that Arcola may have lower grain-Cd accumulation in part because it cycles more Cd back to the roots than Kyle.

PTA062 Allocation of Cd Within *Triticum turgidum* L. var durum as Influenced by Life Stage and Cultivar. Chan, D.Y.* and Hale, B.A., University of Guelph, Guelph, ON, Canada. The objectives of this study were to determine uptake and allocation patterns for Cd in plants already containing environmentally relevant tissue loadings of Cd. Specifically, this study determined the fate of ^{106}Cd delivered, 24h before harvest, to young and mature durum wheat plants. *Triticum turgidum* L. var durum cvs Kyle and Arcola were hydroponically grown to maturity with cadmium nitrate continually supplied to the roots at concentrations of 5.0 or 20.0 µg Cd/L. These cultivars were chosen for potentially contrasting patterns of allocation, as 'Kyle' accumulates much higher concentrations of Cd in the grain compared to 'Arcola'. During three life stages (tillering, flowering and ripening), plants were dosed with ^{106}Cd (in addition to the background dosage with cadmium nitrate) at either 5.0 or 20.0 µg Cd/L. Roots and shoots, as well as heads and grain in the latter life stages were collected 24h after ^{106}Cd dosing. The tissues were then dried and digested using the closed teflon vessel digestion procedure; ^{106}Cd concentrations in the tissue were determined using ICP-MS. The results indicated that Cd was taken up by roots at all three life stages, but is translocated from roots at tillering and flowering stages only; Cd is not translocated from roots to shoots at the ripening stage. Cadmium applied to roots at the tillering stage is translocated to shoots similarly for both cultivars; however, at flowering, only Kyle translocated Cd from the roots to the shoots, and then to flowering heads. These results also demonstrate the utility of stable isotopes in studies of metal movement among biological compartments which already contain equilibrium concentrations of the same metal.

PTA063 Potential of Dandelion (*Taraxacum officinale*) as Bioindicator of Manganese Environmental Contamination. Normandin, L., Université de Montréal, Kennedy, G., École Polytechnique de Montréal, Talbot, D., and Zayed, J., Université de Montréal, Montréal (Québec), Canada. Leaves and roots of dandelion (*Taraxacum officinale*) have previously been shown to be suitable monitors of nickel, cadmium, and lead air/soil pollution. Since the combustion products of methylcyclopentadienyl manganese tricarbonyl (MMT), an antiknock agent used in gasoline, are sources of inorganic manganese (Mn) pollution in the environment, this study aims to evaluate dandelion as a potential bioindicator of Mn contamination. Samples of dandelions were taken at 10, 50 and 100 meters from a highway in Montreal (Quebec) where traffic density is high (135,000 vehicles per day). Soil samples were also taken at the same three sites. Total Mn and other metals (Mg, Ca, Al, Fe and Zn) were determined in plants (stem, leaf, root, flower, and fruit) and soils by neutron activation analysis. Soil pH was also determinated. Mean Mn concentrations in plants were similar for all the sites for each part of the plant (stem: 10.9 - 13.3 ppm; leaf: 23.0 - 32.8 ppm; root: 19.9 - 20.7 ppm; flower: 14.4 - 17.2 ppm; and fruit: 23.1 - 40.5 ppm). In general, metal concentrations decreased in the following order: Ca > Mg > Al > Fe > Zn > Mn in the different structures. Soil Mn, Ca, Fe, and Zn concentrations were significantly higher at sites near the highway (10 m and 50 m) than at 100 m while pH did not vary significantly. Soil Mn mean values (523-768 ppm) ranged within the natural « background » levels which are between 40 and 900 ppm. In conclusion, it appears that the dandelion is not appropriate for monitoring low Mn contamination.

PTA064 Differences in Mercury Concentration Between Chinese and Louisiana Crayfish. L.J. Schuler*, J.P. Howell*, and M.G. Heagler. Department of Biological and Environmental Sciences, McNeese State University, Lake Charles, Louisiana, USA. Mercury is a ubiquitous metal which can be found in high concentrations in aquatic environments. Reports of high concentrations of mercury in fish far from industrial centers have renewed interest in the bioaccumulation of mercury through the food web. The purpose of this study was to examine and compare the mercury concentrations of locally caught and processed crayfish to that of Chinese crayfish. Samples of each type were obtained from area markets, and the abdominal muscle tissue was analyzed for total mercury using CVAAS. Preliminary results show a significant difference (α=0.05) in mercury concentration, with Chinese crayfish averaging 0.059 µg/g wet weight and Louisiana crayfish averaging 0.027 µg/g wet weight. Fresh crayfish were also obtained from area restaurants and analyzed. These results were not significantly different from the locally processed crayfish. The results fall below the action limits established for mercury by both the FDA (1.0 µg/g) and the EPA (0.5 µg/g), but an average meal of crayfish may exceed the reference doses for both men and women.

PTA065 PCB-1268 and Mercury Concentrations in Six Species of Estuarine Fish Exposed to Industrial Sources: Results of Three Biannual Samples. Keller, A.E.,* USEPA, Athens, GA; Francendese, L., USEPA, Atlanta, GA; Manning, R., GAEPD, Athens, GA; Winsness, S., GAEPD, Atlanta, GA ; Meyer, P., USEPA, Athens, GA. Concerns about exposure of the public to contaminants has resulted in the initiation of fish sampling programs in many states. One such monitoring effort has been focused along the Turtle River and its tributaries in southeastern Georgia. The chemicals of concern, PCB-1268 and mercury, emanate from wetlands and sediments adjacent to an industrial site where oil refining, paint and varnish production, and chlor-alkali battery manufacture occurred over a 75 year period. The plant was closed in 1994. Since that time, there has been considerable site remediation including sediment and soil removal, and building demolition. Fish species targeted for collection were red drum, black drum, Atlantic spot, spotted seatrout, striped mullet and sheepshead. Blue crab and white shrimp were also collected at each station. Analyses of 1997 fish samples indicated that consumption advisories in place for two years were still appropriate.

PTA066 Methods of Exposure Assessment: Lead-Contaminated Dust in Philadelphia Schools. Shorten, C.V. and Hooven, M.K., West Chester University, West Chester, PA. The purpose of this study was to develop a method to accurately assess exposure of children to lead in Philadelphia schools. Three wipe sample protocols were developed: one included accessible surfaces such as desktops and windowsills, a second included inaccessible surfaces such as the top of filing cabinets and light fixtures, and a third included hand wipes of the study participants. Because ingestion is the major route of entry of lead into the body, a protocol including wipe samples of accessible surfaces should be a more valid assessment of children's potential exposure than one that includes surfaces which children cannot reach; however, inaccessible surfaces are frequently included in screening assessments. Surface wipes were collected at ten different locations from accessible and inaccessible classroom surfaces (n = 11) and from the palms of student subjects in the same locations (n = 168). One-way ANOVA showed a significant difference in lead dust concentrations determined by the three protocols (F = 4.619; 2,27 df, p = 0.019). In a post-hoc analysis, Tukey's Multiple Comparison Test showed that lead-dust concentrations were significantly elevated at the inaccessible surfaces yet they were uniformly low on the accessible surfaces and the children's palms. The low concentrations found on the accessible surfaces and palms were consistent with observed changes in blood lead levels of study participants. After six months exposure to the study locations, 156 of 168 children experienced no change in blood lead level, while 12 experienced only a minimal change of 1 or 2 µg/dL. The mere presence of lead in inaccessible dust in the school environment does not automatically constitute a health hazard because there may not be a completed exposure pathway.

PTA067 Human Risk Assessment Related to Residual TPH Contamination of a Bioremediated Petroleum Storage Site. Loranger* S., QSAR Inc, Montreal, QC, Canada; Pouliot, Y., Dussault, L., Biogénie Inc, Québec, QC, Canada; Y. Courchesne, QSAR Inc, Trois-Rivières, QC, Canada. From July 1995 to May 1998, an extensive bioremediation program has been conducted on a former petroleum storage site near Quebec city. Several remedial technologies including, air sparging, bioventing, biopile treatment and off-site disposal were used for the decontamination of the impacted soil (40 000 m^3). This program led to a significant reduction of BTEX and PAHs concentrations in the soil and in the groundwater. In spite the reduction of about 80% of the total petroleum hydrocarbon (TPH), soil concentrations exceeded the provincial generic criterion (B) for residential use in some areas of the site. Using the Massachusetts Department of Environmental Protection (MADEP) approach, a human risk assessment was conducted in order to evaluate the impact of this residual contamination on the health of the future resident of the site. This approach is based upon the toxicological evaluation of hydrocarbon fractions (eg. Extractable Petroleum Hydrocarbonn or EPH) and the identification of « reference compounds ». Following the MADEP EPH method, aliphatic (C9-C18, C19-C36) and aromatic (C10-C22) fractions were measured for about 15% of the TPH soil samples (n=30/207). Simple regression indicate that the relation between TPH (independent variable) and other EPH fractions was significantly correlated with r^2 varying between 53% and 72%. A multimedia, multiple pathway exposure model (CalTOX) was used to estimate the environmental and exposure media concentrations, the exposure doses, and the potential risk related to different remedial scenarios. Results show that the overall exposure of the future resident to EPH in soil poses no significant health risk. The sensitivity analysis indicates also that physico-chemical parameters such as K_{oc}, K_{ow} and half-life in soil are the most sensitive.

PTA068 An Integrating Ultraviolet-B Dosimetry System Using Thin-Layer Aluminum Oxide Detectors. Colyott,L.E.*, Oklahoma State University, Stillwater, OK; McKeever, S.W.S., Oklahoma State University, Stillwater, OK; Akselrod, M.S., Oklahoma State University, Stillwater, OK; Bantle, J.A., Oklahoma State University, Stillwater, OK. This presentation describes the development of an integrating ultraviolet-B (UVB) dosimetry system, comprised of UVB sensitive dosimeters and a portable detector readout device. The dosimeters can be reused and the UVB sensitive detectors can be stored until readout is convenient. The UVB sensitive aluminum oxide detectors have no significant temperature dependence in the region of biological interest (0\endash 50 C) and provide a dynamic used in the field or a laboratory setting.

PTA069 Biological early warning systems in drinking water production. Penders, Eric J. and Stoks, Peter G*. WRK Water works, Nieuwegein, The Netherlands. The WRK Water transport company is responsible for the abstraction and partial purification of surface water for the drinking water production of well over 2 million people in The Netherlands. Two water winning stations, one at a side canal of the river Rhine, the other at IJsselmeer lake provide an annual total capacity of 260 Mm^3 / year. In view of the dense population and industrialization upstream (over 20 million people) and the heavy shipping traffic, the control of water quality variations due to, eg. accidental spills, is of paramount importance. An early warning system is, therefore, in operation. In addition to "classical chemistry" such as on-line measurements of temperature, pH, conductivity and turbidity, immunological as well as highly advanced chemical screening techniques are employed. However, using these chemical techniques it is only possible to detect and identify a restricted number of substances. Numerous other pollutants may occur, that cannot be detected and/or that may have unknown toxic properties. For added insight, notably regarding the occurrence of toxic effects resulting from spills, bio-alarming devices have been added. These include on-line fish, mussel and algae monitors as well as light-emitting bacteria. Although there have been several intake stops due to pollutants detected by the chemical early warning system, no stops have yet been based solely on bio-alarms. The responses have, however, been used in assessing the gravity of pollutant levels detected chemically, notably in cases where quality standards or toxicity data could not be obtained.

PTA070 A Retrospective Analysis of the Impact of River Water Quality upon Benthic Fauna. Webb, S.F.*, Procter & Gamble Technical Centres Ltd., Staines, UK; Bird, J.M., Manchester University, UK. The potential impact of surfactants upon benthic fauna relative to other impacts was assessed via a retrospective analysis of a UK Environment Agency database detailing physico-chemical and biological monitoring results from 13 sites on rivers in NW England during 1993-95. Physico-chemical data comprises >30 parameters including MBAS (i.e., correlate of anionic surfactant), monitored weekly or fortnightly. Biological monitoring data include biotic indices (Biological Monitoring Working Party/Average Score Per Taxa) and taxonomic listings to family level. Biological sampling typically took place 3 times/annum. Physico-chemical data was summarised as annual median values for each site. Biological monitoring data was summarised in the form of annualised BMWP and ASPT scores. Bivariate and multivariate statistics were then employed in an attempt to identify physico-chemical and habitat parameters most correlated with biology. Total copper (range 1 - 12 µg/l) was the parameter that best correlated with BMWP ($r = -0.87$) and ASPT scores ($r = -0.86$). Annual median MBAS response at sites was as high as 500 µg/l with individual values >1000 µg/l. Significant correlations of MBAS with BMWP ($r = -0.37$) and ASPT ($r = -0.42$) were observed. Multivariate statistical analysis involved a factor analysis to group the physico-chemical parameters into factors to overcome the high degree of inter-correlation. These factors were then regressed against BMWP ($R^2 = 78\%$) and ASPT ($R^2 = 70\%$) scores. A factor containing total copper, total zinc, total nickel, BOD, alkalinity, phosphate and pH was the best single determinand (assuming causality) of BMWP (33.5%) and ASPT (45.5%) score. A factor that contained MBAS response was low down in the hierarchy of determinant factors for both BMWP (6.8%) and ASPT (7.4%).

PTA071 Using Univariate and Multivariate Techniques to Analyse Community Response to Toxic Perturbation by a Surfactant. Whittle, D.*, Maltby, L., Warren, P.H., University of Sheffield, Sheffield, UK; Tattersfield, L.J., Shell Research and Technology Centre, Thornton, UK. Multispecies model ecosystems have been used to assess the impact of perturbations on aquatic communities. Traditionally such studies have been analysed by separate comparisons of individual variables e.g. using taxa abundance to generate NOECs. An alternative approach is to use multivariate techniques which are capable of providing a measure of the over-all change in community structure resulting from perturbation by taking into account several variables simultaneously. This poster compares the results obtained from three different types of analyses applied to macroinvertebrate abundance data from an outdoor model stream system dosed with an alcohol ethoxylate (a non-ionic surfactant). Responses were assessed in terms of (i) individual species responses, (ii) whole community response in terms of single synthetic variables e.g. diversity indices and (iii) multispecies response using multivariate ordination and clustering techniques. All three analytical approaches are capable of detecting concentration-dependant effects within model communities. At the population level, individual taxa demonstrate considerable variation in their response to perturbation. Diversity measures provide a means of summarising this variation into a single synthetic index of community-level response. Multivariate techniques are the only suite of methods capable of incorporating population-level variation into community-level effect without the subsequent loss of species-specific information.

PTA072 Development of Effects Data for Alcohol Ethoxylate Surfactants Using Stream Mesocosms. Dorn, P. B.*, Shell Chemical Company, Houston, Texas; Tattersfield, L. J., Shell Research and Technology Centre, Thornton, UK; Raney, K. H., Shell Chemical Company, Houston, Texas; Rodgers, J.H., Jr., Clemson University, Pendleton, SC. The aquatic safety assessment of surfactants depends upon the accurate prediction of environmental effects concentrations coupled with exposure assessments. Model stream mesocosms can provide realistic predictions of environmental effects in receiving waters and minimize the need for safety factors if the objectives are properly stated in advance. The ecological effects of a homologous series of four linear-type alcohol ethoxylate surfactants were determined in two stream mesocosm systems located in the US and the UK. The series of homologs ranged between an average carbon chain of C10 to C14.5 with the average ethoxylate distribution between 6 and 9. Sensitivities were evaluated for macrophytic plants, periphyton, invertebrates and fish in series of exposure concentrations. In all of the experiments, the surfactant was accurately delivered and exposure concentrations were not significantly reduced by biodegradation. The mesocosms showed that a factor of 5-25 times decrease in toxicity exists between the C14.5 and the C10 AE's and that fish were generally the most sensitive to surfactant exposures, although some invertebrates such as *Simulium sp.* and *Gammarus* exhibited higher sensitivity in isolated instances. Data from the US and UK systems were similar in spite of their differences in geography, water quality and species. Comparing laboratory data for similar species and groups to the mesocosm results showed that there were typically less than three-fold differences.

PTA073 Critical Body Burdens of the Anionic Surfactant Dodecyl Linear Alkyl Benzene Sulfonate, C_{12}LAS, in Aquatic Organisms. Rawlings, J.M.*, Versteeg, D.J., Procter & Gamble, Cincinnati, OH. Risk assessments for surfactants have traditionally been conducted by interpreting water column concentrations from single species toxicity tests (e.g., LC_{50}). However, effects are expected to be more closely related to internal concentrations (i.e., body burdens) than external concentrations. Critical body burdens (CBB; body burden at lethality) of *Hyalella azteca, Corbicula*, fathead minnow (*Pimephales promelas*) and bluegill (*Lepomis macrochirus*) exposed to the anionic surfactant dodecyl linear alkyl benzene sulfonate (C_{12}LAS) were determined. The organisms were evaluated in acute toxicity, bioconcentration and body burden tests. CBBs were estimated from the LC_{50} multiplied against the bioconcentration factor (BCF) and compared to measured CBBs. Acute 96 h LC_{50} values were 2 - 3 mg/L for *H. azteca*, fathead minnow and bluegill, and 12 mg/L for *Corbicula*. BCFs were variable and were inversely related to exposure concentrations. Estimated CBBs (LC_{50} * BCF) ranged from 1.2 - 3.7 mmoles/kg wet weight. Measured CBBs were less than calculated values for all species and ranged from 0.1 - 1.7 mmoles/kg wet weight. The drop in measured CBBs relative to calculated are due to reduced BCFs at greater exposure concentrations. CBBs within a species varied by a factor of approximately 5. Lipid normalization of LAS concentrations did not change these observations. Hence, whole organism body burdens cannot be calculated from the LC_{50} times the BCF for this surfactant.

PTA074 Periphytic Fate and Effects of Linear Alkylbenzene Sulfonate in a Model Stream Ecosystem. D.M. Lee* and S.E. Belanger, Environmental Science Dept., The Procter and Gamble Co. Cincinnati, OH. In 1996, a comprehensive evaluation was conducted on the periphytic fate and effects of linear alkylbenzene sulfonate (LAS) on periphyton in a model stream ecosystem. Periphyton were colonized on tile substrata for 8 weeks prior to the start of dosing. Streams were dosed with 0-2978 µg LAS/L for an 8-week period followed by a 4-week recovery period. Specific endpoints evaluated included surfactant mineralization, formation of biodegradation intermediates, heterotrophic activity, phytotoxcity, bacterial biomass and community structure. Levels of microbial biomass rose throughout the dosing period and indicated a robust microbial community. Biodegradation, measured as the mineralization of a radiolabeled LAS tracer, indicated that acclimation occurred at LAS concentrations >11µg/L during the dosing period. Acclimation was generally dose dependent and was lost by the end of the 4-week recovery period. Bacterial heterotrophy was unaffected throughout the test by all concentrations of LAS. Levels of algal photosynthesis varied during the dosing period, but were generally higher in the dosed streams versus the control. Community structure, as determined by PLFA analysis, show shifts in the overall community structure associated with LAS dose. Algal taxonomy analysis identified a shift towards increased numbers of blue-green algae by the fourth week of dosing in streams receiving ≥927 µg LAS/L. These results and others will be presented to give an integrated overview of the periphytic community structure and function responses to LAS.

PTA075 Toxicity of Mixed Surfactant Systems. Russell, G.L.* and Britton, L.N., CONDEA Vista Co., Austin, TX. Since cleaning products are generally composed of mixtures of surfactants and since their release into environmental compartments is often above the critical micelle concentration (cmc), it is important to understand the toxicity of these mixed systems. Toxicity can be viewed as additive when surfactant mixtures are below the cmc; howerver, above the cmc surfactants form mixed micelles and may elicit an unpredictable response on target organisms. Microtox™ assays were conducted on linear alcohol ethoxylate (AE), linear alkylbenzene sulfonate (LAS), alcohol sulfate (AS), alcohol ether sulfacte (AES) and α olefin sulfonate (AOS) surfactants. Large molecular weight AE are relatively non-toxic when compared to the anionic surfactants, and these large AE have the ability to reduce the toxicity of AE mixtures containing LAS but not the other anionics. Surface tension measurements suggest that the mechanism of toxicity reduction is the decrease in LAS monomer concentration in large AE:LAS mixtures by decreasing the cmc of the surfactant mixture. An intriguing, potential application of this knowledge is the addition of surfactants, such as the large AE, to cleaning products in order to reduce the toxicity of mixtures containing desirably performing yet moderately toxic surfactants.

PTA076 Association of Alcohol Ethoxylate (AE) Surfactants with Dissolved Humic Substances. Kerr, K.M.*, and McAvoy, D.C., The Procter & Gamble Company, Cincinnati, Ohio. The environmental fate, transport, and toxicity of surfactants in the environment are strongly controlled by their surface active properties. In this study we investigated the association of alcohol ethoxylate (AE), a non-ionic surfactant, with a dissolved humic substance using a reverse-phase C 18 Sep-Pak column. Purified radiolabelled homologues of AE were synthesized at Procter & Gamble Company, whereas the model humic substance was obtained from Aldrich and purified at The Ohio State University. Results showed that the association of AE with dissolved humic substance increased with an increase in alkyl chain length with little effect due to changes in ethoxylate group number, suggesting that a hydrophobic mechanism of association dominates this interaction. These results also indicate that on an equivalent molar basis the shorter alkyl chain length AE surfactants would be more available in solution.

PTA077 Effect of Ethoxylate Number and Chain Length on the Biodegradation Kinetics and Sorption of Linear Alkyl Ethoxylates in Activated Sludge. Itrich, N. R., Kerr, K. M, McAvoy, D. C., and Federle, T. W.*. The Procter and Gamble Company, Cincinnati, OH. A series of batch activated sludge die-away studies and sorption experiments were conducted to determine the first-order biodegradation rates and sorption coefficients of various homologs of alkyl ethoxylate. 14C-(ethoxylate) C14E1, C14E3, C14E6 and C14E9 were used to investigate the effect of EO number, while 14C-(ethoxylate) C12E6, C14E6 and C16E6 were used to examine chain length effects on biodegradation and sorption. Die-away studies consisted of dosing a trace level (0.2 uM) of each homolog into fresly collected activated sludge and monitoring the disappearance of parent, formation of metabolites, formation of 14CO2 and incorporation into solids with time. EO number had little effect on the first order decay rates for primary biodegradation, which equaled 68-78 hr-1. However, the C16 chain length homolog exhibited a slower rate of parent decay (39 hr-1) compared to its corresponding C12 and C14 homologs (65-66 hr-1). With the exception of C14E1, biodegradation occurred via central cleavage yielding the corresponding fatty alcohol and PEG group. In the case of C14E1, biodegradation involved both central cleavage and omega oxidation of the terminal ethoxylate alcohol. Mineralization rates ranged from 23 to 46 hr-1 with the slowest rate exhibited by C16E6 and the fastest rate by C14E9. Sorption coefficient (Kd) tended to decrease with EO number and increase with chain length with the latter having the more profound effect. Parameterization of wastewater treatment models with these data indicates high removal (> 99.3%) of all homologs and > 85% removal of PEG metabolites. Chain length rather than ethoxylate number seemed to have the greatest affect on the biodegradation rate with the most sorptive homolog exhibiting the slowest biodegradation rate. However, modeling indicates that this slower rate would not adversely affect removal since its offset by higher sorption removal.

PTA079 Measuring the Biodegradability of C8- and C-9 Alkylphenol Ethoxylates and their Biodegradation Intermediates. Staples, C.A.*, Assessment Technologies, Inc., Fairfax, VA; Williams, J.B., Union Carbide Corporation, South Charleston, WV; Naylor, C.G., Huntsman Chemical Corporation, Austin, TX. A series of biodegradation tests was conducted using nonylphenol ethoxylates (NPE-9), nonylphenol ether carboxylates (NPEC-1,2), octylphenol ether carboxylates (OPEC-1,2), the low mole ethoxylates (NPE-1,2), and nonylphenol. Test methods used were developed by the Organization for Economic Cooperation and Development (OECD). OECD method 301B (modified Sturm test measuring ultimate biodegradation to CO_2 was used for most tests, while OECD method 301F (manometric respirometry, measures oxygen consumption) was used with nonylphenol. To be considered readily biodegradable based on these tests, a compound must achieve >60% theoretical carbon dioxide formation ($thCO_2$, method 301B) or theoretical oxygen consumption (thO_2, method 301F) in 28 days and achieve the 60% within 10 days of achieving the first 10% (also called the ten day window). Test results achieving between 20 to 60% $thCO_2$ or thO_2 formation, are designated inherently biodegradable. Tests achieving <20%$thCO_2$ or thO_2 formation are considered persistent. The results for all compounds tested, including the commercial product and their biodegradation intermediates, show that all are readily or inherently biodegradable. For some compounds showing inherent biodegradability (e.g., nonylphenol), a sufficient extent of biodegradation occurred (>60%), but not within the specified time frame. These standard and stringent laboratory test results are discussed in context with their use and extrapolation to field situations. Finally, these new data will be used with data from previous studies to better establish the biodegradation pathways of alkylphenol ethoxylates.

PTA080 An Environmental Risk Assessment of Nonylphenol Ethoxylates and their Biodegradation Intermediates in Water and Sediments. Staples, C.A.*, Assessment Technologies, Inc., Fairfax, VA; Williams, J.B., Union Carbide Corporation, South Charleston, WV; Naylor, C.G., Huntsman Chemical Corporation, Austin, TX. An environmental risk assessment of nonylphenol ethoxylates (NPEs) and their biodegradation intermediates was conducted for surface waters and sediments. Environmental risk was calculated using comparisons of predicted exposure concentrations (PECs) and predicted no effect concentrations (PNECs). A PEC / PNEC ratio of <1 is desired and indicates no environmental concern. The compounds examined were commercially relevant products (e.g., NPE-9), and their biodegradation intermediates (mainly low mole ethoxylates, NPE-1,2; low mole ether carboxylates, NPEC-1,2; and occasionally nonylphenol). NPEs are ultimately biodegradable in surface waters, but are often detected near the location of sewage treatment plant outfalls due to their wide use in consumer products. Examination of the potential risks caused by exposures to NPEs and their biodegradation intermediates is warranted. Exposure concentrations were obtained from measurements of surface water and sediment concentrations in North American waters and were used to estimate PECs. Sufficient aquatic toxicity data are available to calculate a PNEC for the most acutely and chronically toxic of the biodegradation intermediates, NP. Some data are available for the other degradation intermediates of NPEs, which were used to calculate PNECs for those compounds proportional to that of NP. PEC / PNEC ratios were calculated for in-stream water and sediment concentrations (95^{th} percentiles and maximum concentrations) of these compounds. Based on 95^{th} percentile surface water and sediment concentrations, all PEC / PNEC ratios were less than one. Based on maximum concentrations, PEC / PNEC ratios ranged from much less than one to a high >0.1 for available in-stream water concentrations and from much less than one to a high of about one for sediments.

PTA080A Recent Residue Levels of Pesticide and Transformation Products in Mollusks of US Coastal Waters and the Great Lakes. Johnson, W.E. and O'Connor, T.P. NOAA, National Ocean Service, Silver Spring, MD. The National Oceanic and Atmospheric Administration's (NOAA) National Status and Trends Program (NS&T)

monitors nationwide levels of toxic chemicals in fish, shellfish, and sediments. Included in the nearly 80 chemicals monitored at over 240 sites since 1986 are fifteen banned or limited use pesticides including DDT, its breakdown products DDD and DDE, and nine other chlorinated pesticides. Starting in 1993 additional pesticides were added to the list of compounds analyzed and included several current-use pesticides and additional transformation products of the chlorinated pesticides (chlorpyrifos; endosulfan; αHCH, βHCH, and δHCH isomers of lindane; α-chlordane, γ-chlordane, oxychlordane, and cis-nonachlor). Mean chlorpyrifos levels in zebra mussels (dry-wt) from the first year of monitoring (1995-6) ranged from 3.5 ng/g to 21.8 ng/g with an apparent decreasing concentration trend with increasing distance downstream from Lake Michigan to Lake Ontario. Ratios of metabolite to parent compound were followed as a function of year for several compounds. The ratio of DDE to DDT showed an increasing trend from 1984 to 1995. Similarly ratios of the α, β, δ HCH isomers of lindane also showed increasing trends over the four year period (1994-1996). In general, the mean concentration of the parent compounds show a decreasing trend and initial indications suggest an increasing trend in concentration of transformation products.

PTA081 Bioaccumulation of organotin compounds by polychaete *Perinereis nuntia*. Ikeda, K.* and Kakuno, A., National Research Institute of Fisheries Science Kanagawa, Japan. Organotin compounds have been detected mainly in sediment in coastal environments and seem to bioaccumulate by benthic organisms. Benthic organisms play an important role in transfer of organotin compounds from sediment to a benthic food chain. Therefore, the bioaccumulation of organotin compounds from seawater and sediment and also their elimination were studied by an eight weeks exposure experiment using polychaete *Perinereis nuntia*. Its bioconcentration factors (BCF) of TBT and TPT were 1,000 and 16,000, respectively and its biomagnification factor (BMF) of TBT from sediment was 1. Its elimination rate constant of TBT, 0.05/day, was independent on the uptake routes in polychaete. Detecting metabolites of TBT in polychaete, it suggests that polychaete can metabolite TBT and its metabolic pathway is similar to that of a red sea bream, a marine fish. The TBT elimination rate constant of polychaete seemed to be higher than those of other invertebrate and similar to that of red sea bream. In the case of polychaete, the estimated BCF and BMF for TBT and the actually determined TBT concentration in the Tokyo Bay seawater and sediment suggest that TBT in the sediment has higher contribution on the bioaccumulation of TBT by polychaete than that of the seawater.

PTA082 Extrapolation in Human Health and Ecological Risk Assessments. Munns, W.R., Jr., US EPA, Narragansett, RI, MacPhail, R.C.*, US EPA, RTP, NC, Erickson, R.J., US EPA, Duluth, MN, Harris, T.S.,US EPA, RTP, NC, Lackey, R.T., US EPA, Corvallis, OR, Luebke, R.W., US EPA, RTP, NC, Rogers, J.E., US EPA, Gulf Breeze, FL, Waters, M.D.,US EPA, RTP, NC. Extrapolation lies at the core of human health and ecological risk assessment. All risk-based environmental decisions are based upon extrapolations of measured endpoints to effects of concern to society. Such extrapolations are a major source of uncertainty in human health and ecological risk assessments. The Fourth National Health and Environmental Effects Research Laboratory (NHEERL) Symposium on Research Advances in Risk Assessment (April 27-30, 1998, Cary, NC) drew upon international participation to focus on key research issues as well as the methods, models, and data employed to extrapolate through temporal and spatial scales and across biological organization. The fundamental objective of the symposium was to explore approaches for reducing uncertainties in human health and ecological risk assessments. Presentations and panel discussions identified commonalities and differences in health and ecological research approaches and extrapolation procedures. Sessions addressed major uncertainties associated with extrapolations among taxa and across biological and ecological organizational hierarchy, through time, and across spatial scale. The concluding session attempted to identify mutually beneficial approaches utilized in health and ecological risk assessment research and decision making. It was recognized that: (1) research that is useful in risk assessment is both scientifically credible and relevant in its policy implications, (2) the evaluation of ecological health and integrity extends beyond traditional risk assessment paradigms and incorporates societal values and preferences, and (3) some decisions regarding ecological risks ultimately also influence human health and survival, but ecological benefits may or may not coincide with human health benefits.

PTA083 Stream Microcosm studies of Zinc Accumulation in Freshwater Benthic Periphyton and Grazing Invertebrates. C. Harris*, L. Bendell.-Young, Department of Biological Sciences, Simon Fraser University, Burnaby, B.C. V5A 1S6. Mining effluent is a complex mixture which can contain high levels of both dissolved and particulate iron (oxides). Stream microcosm studies were designed to evaluate the bioaccumulation of Zn by periphyton and snails in the presence and absence of iron oxides. Six flow through streams (35 L capacity) were constructed and fed with chilled dechlorinated water (1.3 L/min). Current (5-30 cm/sec) was generated via a speed controlled electric motor turning 6 PVC paddlewheels. To ensure periphyton growth overhead light banks provided full spectral light (~20ulux/m2/sec). Treatments were delivered into all streams via a multi-channel peristaltic pump. Experimental design was 2*2*2 with treatments consisting of control, 50ug Zn/L with no Fe and 50ug Zn/L with 200ug Fe/L. To establish initial conditions, periphyton was cultured by recirculating nutrient rich water within the microcosm for a two month period. Once treatments commenced (t = 0) the system switched to complete flow through. Gastropods were introduced at 10 days. The streams were repeatedly sampled over the 30 day duration for periphyton biomass, chlorophyll and concentrations of Zn in the water, periphyton and gastropods. Biomass and pigment analysis varied over time, but no significant differences were observed among streams indicating that treatments did not cause toxic effects. In treatments receiving high Fe, Zn uptake by periphyton and gastropods was reduced. Mean levels of Zn in the periphyton were found to be 130, 460 and 1530 ug/g in the controls, Zn/Fe and Zn treatments respectively. Tissue Zn levels in the grazing gastropods correlated (r >0.80) with the periphyton Zn concentrations in all streams except those receiving the Fe treatments. These findings indicate that periphyton would be an excellent monitoring tool and appears to be highly sensitive to changes in overlying water chemistry.

PTA084 Relation between Cd Concentrations in the Biomonitor *Chaoborus* and those of its Planktonic Prey. Croteau, M.-N.*; Hare, L., INRS-Eau, Sainte-Foy, Québec, Canada. Cadmium concentrations in a lake-dwelling insect (*Chaoborus*) can be related to those in their habitat with the free-ion activity model, provided that competition between hydrogen and cadmium ions for biological uptake sites is taken into account. The strong relationship observed between Cd concentrations in *Chaoborus* larvae and free Cd ion concentrations does not necessarily mean that the larvae take up their Cd directly from water. Recent information indicates that Cd accumulated by the predator comes almost entirely from its prey. We tested the hypothesis that Cd concentrations in *Chaoborus* are directly related to those in its planctonic prey by measuring Cd concentrations in the predator and in prey collected from a large number of lakes. We show that Cd concentrations in *Chaoborus* larvae are not accurately predicted by Cd concentrations in bulk zooplankton samples. A better relationship was obtained when potential prey were separated according to type, that is, cladocerans or copepods. Our results suggest that biological variables such as prey type, ingestion rate and Cd assimilation efficiency influence Cd concentrations in the predator.

PTA085 Food versus water as cadmium sources for the insect biomonitor *Sialis* (Megaloptera). Roy, I.*,and Hare, L. INRS-Eau, Université du Québec, Sainte-Foy, Québec, Canada. Measurements of metal concentrations in animals can be used to class lakes according to their contamination level in a way that is biologically meaningful. The alderfly *Sialis* is a promising biomonitor candidate because of its wide distribution, large size and ease of maintenance in the laboratory. Using such an animal as a metal biomonitor depends on establishing a direct relationship between its metal concentration and that of metal in its surroundings. An important step towards this end is to determine from which environmental compartment the animal obtains its metal, that is, from water or from its food. We exposed *Sialis* larvae in the laboratory to cadmium (Cd) either in water alone, in food alone, or via both food and water (assuming that Cd uptake by the two exposure routes is additive). Our results indicate that the animal takes up Cd from both water and its food (other invertebrates). Because food was an important Cd source, we measured a direct relationship between Cd concentrations in prey and those in the predator. Our results suggest that differences in prey community composition among lakes could influence predator Cd concentrations.

PTA086 Mercury in a Mississippi Reservoir: Sediment and Water Contamination Associated with Fish Consumption Advisories. Allgood, J.C.*, Steevens, J.A., Lutken, C.B., Grace, C.A., Buchannon, R.C., Swann, C.T., and Benson, W.H. The University of Mississippi, University, MS. Mercury has become one of the leading chemicals of concern that have the potential to affect human health and environmental quality in the Southeastern United States. Enid Lake, a U.S. Army Corps of Engineers flood control reservoir located in North Mississippi presently has a fish consumption advisory for mercury. In 1995, a survey of mercury concentration in fish tissue from Enid Lake, found Largemouth Bass contained levels of mercury exceeding the Food and Drug Administration action level of 1 ppm. The lake encompasses a total area of 44,036 acres of land and water, with a permanent pool of 8,400 acres and a flood pool of 28,000 acres. Public use is extensive, with 35,900 acres for recreational use including 458 campsites, 13 boat ramps, and 5 public swimming beaches. The present study was conducted to increase our current understanding of mercury contamination in the reservoir system. Mercury concentrations in water and sediment of the Enid watershed were measured to assess the potential for mercury exposure to the local human and wildlife population. Eight transects of the area, six north/south and two east/west, were selected and samples collected along these transects. A total of 88 water and 44 hydrosoil samples, plus QA/QC samples, were analyzed for mercury. In addition to mercury analysis, water chemistry data including pH, dissolved oxygen, depth, temperature, redox potential, total dissolved solids, and specific conductance were determined. Mercury concentrations in sediment ranged from 0.067 to 0.692 ppm and 0.062 ppb to 0.602 ppb for water. While the total concentration of mercury in water and sediment is indicative of mercury exposure to human and wildlife populations, other water chemistry factors may also influence the bioavailablility.

PTA087 Mercury Contamination in Abiotic and Biotic Compartments of the North Fork Holston River, VA and TN. Dye, S.K.*, Benton, M.J., East Tennessee State University, Johnson City, TN. A survey of mercury contamination in several abiotic and biotic compartments of the North Fork Holston River (NFHR) was conducted. The NFHR flows 217 km from southwest Virginia to northwest Tennessee. Industrial activity of a now-defunct chloralkali plant at Saltville, VA, from 1951 to 1971, resulted in severe mercury contamination of the NFHR. Abiotic (water, sediment) and biotic (periphyton, snails, fish) samples were collected from 17 stations along the entire course of the NFHR, and analyzed for total mercury by cold vapor atomic absorption spectrophotometry. Mercury levels in all water samples was below detection limit (BDL). In sediments, mercury concentrations were 7.29 µg/g at Saltville, BDL upstream, and 0.456 µg/g downstream. Mean mercury concentrations in periphyton were BDL upstream, 0.374 µg/g downstream. Whole-body snail tissue mercury concentrations were 0.098 µg/g at Saltville, BDL upstream, and 0.228 µg/g downstream. Mean whole-body fish tissue mercury concentrations were 0.201 µg/g at Saltville, 0.060 µg/g upstream, 0.369 µg/g downstream. Results show that mercury in the NFHR ecosystem continues to cause severe contamination in all measured compartment at the original impact site at Saltville. Moreover, contamination has spread significantly downstream in all measured compartments. Upstream sites remain largely unaffected.

PTA088 Mercury Concentrations in Food. Holt, B.J.*, M.G. Heagler, McNeese State University, Lake Charles, LA. Generally people consider the food purchased from their supermarket to be free from possible contamination, but this may not always be the case. The U.S. Food and Drug Administration (FDA) oversees the products found in our supermarkets, including fruits and vegetables, meat, poultry and fish. Measurements will be made to see if some of these foods contain unusually high concentrations of mercury. Researchers have measured mercury concentrations of raw fish tissue, and some types of fish, including swordfish (1.19 μg Hg/g ± 0.80), exceed the FDA's action level of 1.0 μg of mercury/g of tissue. However, the handling and cooking of the fish prior to consumption may modify the mercury concentrations. Therefore, measurements were made to compare the mercury concentrations of fish, specifically swordfish, before and after cooking. Data suggests that there is no statistically significant change in the mean concentration between the raw swordfish, which was 0.642 μg Hg/g (wet weight), and the cooked swordfish, which was 0.648 μg Hg/g (wet weight). Measurements will continue to be made with variations in the method of cooking, and different varieties of fish.

PTA089 A Comparison of Human Health Risk with Risk to Wildlife for Nitroaromatic Munition Compounds. Talmage, S.S.*, Opresko, D.M., Hovatter, P.S. Oak Ridge National Laboratory, Oak Ridge, TN. Both U.S. Environmental Protection Agency Reference Doses (RfDs) to protect human health and screening benchmarks to protect mammalian wildlife are usually extrapolated from feeding studies utilizing laboratory animals. Screening benchmarks calculated for six nitroaromatic munition or munition-related compounds (2,4,6-trinitrotoluene, 1,3,5-trinitrobenzene, 1,3-dinitrobenzene, RDX, HMX, and tetryl) were compared to human RfDs. All values were based on NOAELs for the test species. The use of different endpoints for the two types of values and the necessity for application of an interspecies uncertainty factor (usually 10) for extrapolation from animals to humans, lowered human RfDs below wildlife screening benchmarks by a factor of 60 (HMX) to 3000 (2,4,6-trinitrotoluene). However, for wildlife, laboratory test NOAELs are scaled on the basis of differences in body size instead of using the interspecies uncertainty factor. Endpoints for wildlife were based on population-level effects, usually testicular effects, whereas those for human health were based on biochemical/organ weight changes or lesions. The conservativeness of both values was related to the availability of chronic studies as well as the chosen test species. Data from a series of studies with 1,3,5-trinitrobenzene also indicate that wildlife species may be less sensitive to toxicants than the laboratory rat or mouse.

PTA090 The Significance of Food in Decreasing the Influence of Environmental Loads on Human Organism. T.F.Chirkina, S.D.Zhamsaranova, Z.A.Plastinina, ESSUT, Russia, Republic of Buryatia, Ulan-Ude.1.The significance of food as the most important recovery regulator of the broken endo/ecological balance is increasing under the present conditions of growing biospheric overloading. 2. The influence of unfavourable factors on human organism leads to endo/ecological autointoxication revealing itself in the immune system relaxation of the self-defence mechanism. 3. The methods of immune correction of human organism systems are being developed at the East-Siberian State University of Technology by working out some special food products during this decade. 4.Such an approach is based on the fact that biologically active substances in food do not react so vividly as in pure form. Food diet can be used for the sake of preventive measures for secondary immune deficiencies . 5. We have proved that spleen, thymus and bone marrow(red) of slaughter cattle as well as wild grassy plants of Buryatia can be used as food stimulators for immune system. 6. Technological recommendations for their use in food products in the form of extracts, supernathanls and hemogenants have been done.

PTA091 Fish and Fishing: Potential Hazards to Fish and Fishermen. Burger, J., Gochfeld, M. G. Environmental and Occupational Health Sciences Institute, Piscataway, NJ, Gaines, K.F., Boring, C.S. Savannah River Ecology Laboratory, Aiken, SC. Survey data obtained from fishermen along the Savannah River between Georgia and South Carolina indicates that a substantial number of fishermen and their families are eating more than the risk assessment assumption of 19 Kg/year. There are consumption advisories for the Savannah River, based mainly on mercury. In this study we examine the levels of radiocesium and mercury in seven species of fish collected from the Savannah River. Fish were selected for collection based on trophic level, and included some of the common fish eaten by the local fishermen. Mean mercury levels were above the allowable 1 ppm for large mouth bass, and approached this level for sunfish and black crappie. Radiocesium levels varied significantly by site, with levels being significantly higher along the Savannah River Site, particularly for bowfin and large mouth bass. We discuss the health implications for both fish communities and human fish consumers. Work supported by CRESP through the Department of Energy, AI#DE-FCo1-95EW55084.

PTA092 **Mercury concentrations in brook trout and white perch from Kejimkujik National Park, Nova Scotia.** d'Entremont, A., d'Entremont Environmental Solutions, Hammonds Plains, NS; Carter, J.A., Carter Environmental Consulting, Halifax, NS; Burgess, N.M.*, Canadian Wildlife Service, Sackville, NB; Drysdale, C., Parks Canada, Kejimkujik National Park, NS; Brun, G., Environment Canada, Moncton, NB. In recent years, mercury concentrations in larger freshwater sportfish collected from lakes in the Canadian Maritime Provinces have been reported to exceed the Health Canada safe consumption guideline, resulting in the issuance of provincial consumption advisories to the public. This study examined fillet mercury levels in brook trout (*Salvelinus fontinalis*) and white perch (*Morone americana*) collected from water bodies at Kejimkujik National Park, Nova Scotia, Canada. These species are important components of a local recreational fishery. Total mercury concentrations ranged from 0.06 to 1.37 µg/g in 81 brook trout with a mean fork length of 26 cm, weight of 264 g and age of 3.3 years. In 30 white perch, total mercury concentrations ranged from 0.35 to 2.30 µg/g. The white perch collected were larger (mean fork length 29 cm, weight 399 g) and older (mean age 9.8 years) than the brook trout. These mercury levels are similar to those observed in other studies in northeastern North America and the Great Lakes area. Fifteen percent of the brook trout and 93% of the white perch samples exceeded the Health Canada safe consumption guideline of 0.5 ppm mercury. Significant positive correlations were identified between total mercury concentrations and fish age, length and weight in both species. Differences in mean mercury concentrations between sampling locations were also statistically significant. Fish methylmercury levels and relationships with local water chemistry will also be discussed.

PTA093 **Intake of Polychlorinated Biphenyls from Food in Italy.** Zuccato, E.*, Mariani, G., Mangiapan, S., Grasso, P., Fanelli, R., Istituto di Ricerche Farmacologiche Mario Negri, Milan, Italy. The dietary intake of total and congener-specific polychlorinated biphenyls (PCB) was determined by the duplicate portion method in 20 volunteers eating a typical Italian diet, i.e. a diet for which no previous data were available. On three non-consecutive days, the volunteers set aside matched portions of all the meals and snacks eaten daily, giving three separate 24-hour collections for each volunteer. After extraction and purification, samples were subsequently analyzed for their PCB content by GC-MS. The mean±SD intake of total PCB was 3.72±1.51 µg/person/day (range 0.97-10.59 µg/person/day). IUPAC congeners 153, 18, 138, 101, 52 and 28 were the PCB found in the highest concentrations, the non-*ortho* coplanar congeners 77, 126 and 169 being globally present as 0.5% of the total. The corresponding levels of toxicity (TCDD toxic equivalency -TEQ- values ascribable to PCB) were within the range of 5-120 pg/person/day of TCDD-equivalents in 18 subjects, i.e. presumed low-risk levels, but were substantially higher (2109 and 4553 pg/person/day) in two subjects with significant intakes of the congener 126. This supports the published evidences that PCB contamination of food might well be a risk for human health. Principal component analysis and redundancy analysis were used to identify the principal sources of PCB in the diet. Dairy products, meat and fish were the main sources, while vegetables contained the most toxic PCB congeners and were the main contributors to total TEQ ascribable to PCB.

PTA094 **The Influence of Diet on Persistent Organic Pollutants in Blood Plasma from Russian and Norwegian Arctic Populations.** Burkow, I.C.*, Norwegian Institute for Air Research, Tromsø, Norway, Odland, J.Ø., University of Tromsø, Tromsø, Norway, Klopov, V., Regional Centre *Monitoring of the Arctic*, St.Petersburg, Russia, and Lund, E., University of Tromsø, Tromsø, Norway. Socio-economic conditions, health services, culture, individual lifestyles, genetics, and environmental contaminants are factors contributing towards health and illness of the Arctic population. As a part of the Arctic Monitoring and Assessment Programme (AMAP), a Russian-Norwegian co-operation project was established to assess the exposure of humans to persistent organic pollutants (POPs) in Arctic areas of Russia and Norway. Concentrations of chlorinated pesticides and polychlorinated biphenyls (PCBs) in blood and breast milk samples were determined by high resolution gas chromatography with electron capture detection and mass spectrometry. The POP levels in maternal plasma among the non-indigenous women in western Siberia were higher than the native population, especially in total PCB, hexachlorocyclohexanes and the DDT-group. The values were higher than the levels reported for the Scandinavian countries, and up to levels of medical concern. The dietary questionnaires showed that the non-indigenous populations of Yamal and Taymir (Russia) consumed considerably less local food items. The global transport, which is clearly shown in studies from Greenland and Canada, can not be the only explanation for the high contaminant levels found. The most important sources of persistent organic pollutants for the Russian Arctic populations of western Siberia seems to be imported food from other areas of Russia and local use of pesticides.

PTA095 **Case Study: Quantifying Fish Ingestion Risks Associated with Food Chain Transfers of PCBs.** Rosenstein, A.B.* and Howell, C.H., Arthur D. Little, Inc., Cambridge, Massachusetts. Ingestion of contaminated fish from the Souhegan River in Milford, New Hampshire, by recreational anglers is a potential exposure pathway for contaminants originating from the Fletcher's Paint Superfund Site. PCBs are of particular concern at this site. Prior to the collection of fish samples, as a first step in assessing this pathway, a screening approach was adopted in 1994. The screening approach predicted upper bound fish tissue concentrations of contaminants using conservative models based on Souhegan River sediment contaminant concentrations. A comparison of modeled fish tissue concentrations to human health levels of concern indicated that contaminants could potentially bioaccumulate in two edible species of fish above levels of concern. To confirm modeled predictions and to better evaluate human health and ecological risks, a sampling and analysis program of Souhegan River fish, focusing on PCBs and pesticides, was completed. The model had significantly underestimated the PCB body burdens in the Souhegan River fish. Risks were calculated for ingestion of upstream and downstream fish by recreational angler. Both average and upper bound exposure assumptions were used. Estimated incremental cancer risks downstream were only slightly higher than the estimated upstream cancer risks, possibly due to upstream contributors of contaminants. All fish ingestion risks were within the generally accepted U.S. EPA guidelines.

PTA096 **Food-Chain Transfer of Incinerator Emissions to Humans and other Species.** Prann, R.S.* C.M. I, and D. McKean, IT Corporation, Somerset, NJ. Food-chain transfer of incinerator emissions were evaluated in the vicinity of a hazardous waste incinerator located in Missouri. Transfer to humans through inhalation, ingestion of soil, surface water, beef, milk, chicken, eggs, and fish was quantified using USEPA guidance for indirect exposure assessments. Although this methodology has been rigorously refined over the past several years, transfer of incinerator emissions to ecological receptors has been relatively un-studied. The methodologies used to calculate the fate and transport of incinerator emissions for human health risk assessment are easily applied to incorporate ecological receptors and food-chain interactions. A case study of the human health food-chain transfer of incinerator emissions is presented and a theoretical ecological food-chain model is also presented.

PTA097 **Pentachlorophenol as a Predicator of Dioxin Content of Wood in Livestock Facilities.** Fries, G.F.*, U.S. Department of Agriculture, Beltsville, MD; Feil, V.J., Zaylskie, R.G., U.S. Department of Agriculture, Fargo, ND; Bialek, K.M., Rice, C. P., U.S. Department of Agriculture, Beltsville, MD. A survey of polychlorinated dibenzo-*p*-dioxin (PCDD) and dibenzofuran (PCDF) residues in beef identified several facilities that yielded samples with higher concentrations than normal background. Analysis of environmental samples from the sites suggested that the high PCDD/F concentrations were associated with the presence of pentachlorophenol (PCP) treated wood in the animal facilities. Our purpose was to determine if PCP analysis provided an economical method to identify wood with high concentrations of PCDD/Fs. Both PCP and PCDD/F analyses were available for 40 samples of wood and other materials collected from 7 facilities. Concentrations of PCDD/Fs as toxic equivalents (TEQs) in non-wood samples were 1 to 3 pg/g while concentrations of PCP were below the quantitation limit. Concentrations of PCP in wood were in the range < 1 µg/g to 8,500 µg/g, and total TEQs were in the range of 10 to 320,000 pg/g. The PCDD/F residues generally were composed of 85 to 95% PCDDs and 5 to 15% PCDFs. The R^2 of linear regressions of the log concentrations of individual congeners and total TEQs on log concentrations of PCP were in the range of 0.75 to 0.80. We conclude that PCP analysis is a useful screening technique for estimating PCDD/F concentrations in treated wood, and that animal access to high residue wood may be an important route of entry of PCDD/Fs to human foods.

PTA098 Chemical Specific Sediment Concentrations for Protection of Benthic Organisms and Fish Consumers. Keating, F.J.*, U.S. EPA, Washington, DC; Armitage, T.M. U.S. EPA, Washington, DC; Berry, W.J., U.S. EPA, Narragansett, RI; Mount, D.R., U.S. EPA, Duluth, MN; Peters, E.C., Tetra Tech, Fairfax, VA. Using equilibrium partitioning theory, interstitial water concentrations can be reliably predicted for nonionic organic compounds from a sediment's organic carbon content and the compound's organic carbon partition coefficient (K_{oc}). Applying this theory, we can associate sediment concentrations, expressed as chemical mass per unit sediment organic carbon mass, with any chosen interstitial water biological effects concentration. By setting interstitial water concentrations equal to final chronic values (FCV) or secondary chronic values (SCV), as defined in EPA guidance, Sediment Quality Advisory Levels (SQAL) have been calculated for 34 individual nonionic organic compounds. They are intended to protect 95 percent of benthic dwelling organisms from the direct effects of exposure to these compounds. SQALs are generally higher than sediment quality guidelines derived empirically from field data containing chemical mixtures. SQALs are best suited for applications which demand a clear link between sediment concentrations of a specific chemical and its capacity to cause sublethal toxicity. The U.S. EPA has used these SQALs to screen sediment chemistry monitoring data and identify where adverse effects on benthic communities may be occurring. In addition, sediment chemistry screening values for the protection of human and wildlife fish consumers have been derived from chemical-specific fish tissue risk levels, representative fish lipid content of the portion consumed, sediment organic carbon content, and frequency distributions of field measured biota-sediment accumulation factors (BSAF) by nonionic organic chemical class. SQALs and sediment chemistry screening values for fish consumers will be plotted on national frequency distributions of ambient monitoring data for specific compounds to show how often these values are exceeded.

PTA099 Environmental impact of pesticides in India and its management through eco-compatible pesticide formulations. Jaglan, R.S., Dept. Entomol.,CCS HAU, Hisar, India; Arora, R.K., SKUAST, RHRS, Udheywalla, Jammu Tawi, India. Pesticide usage in India increased when high yielding varieties responsive to fertilizers and irrigation were introduced. India has trebled its pesticide consumption at a rate of 20 per cent per annum i.e. about 450 g per ha, much lesser than the developed countries. Every year agricultural losses due to pests and diseases amount to Rs.20000 crore and exports more than Rs.1000 crore are rejected owing to poor quality. Alarming resistance in pests and diseases and resurgence of minor pests as major pests have aided in aggravating the losses. Studies on market samples of fruits and vegetables, meat, fish, milk and milk products showed high levels of pesticide residues. Farmers, use highly toxic pesticides on vegetable crops just before harvesting and dip vegetables in pesticides solution. The unrestrained use of pesticides kill plant-friendly parasites, predators and pollinators, thus causing an imbalance in their habitats. The domain of eco-oriented sustainable environment is nourished by a number of multidimensional inputs including those of indispensable agriculural chemicals. The search for more potent and relatively safer pesticides effective at a lower median dose may be accepted on an eco-compatible concept. Since newer pesticides may not be readily reckoned with, development of eco-compatible formulations may be suitably capitalized for integrated pest management which can widen the range of bio-efficacy with minimum residues and related hazards. Pesticide pollution must be diluted by the use of, eco- compatible bio-pesticides. The concept of eco-compatibility, basically involving economic threshold and injury level of the pest requires extensive appraisal by the farmers in India. The concept of IPM through eco-compatible practices involving bio pesticide formulations should provide suitable guidelines for sustainable environment.

PTA100 Trends and Patterns in Chlorinated Hydrocarbon Contaminants in Osprey from the Pacific Northwest. L. Wilson*, and J. Elliott, Canadian Wildlife Service, Delta BC, M. Machmer, Pandion Ecological Research Ltd., Ymir, BC, C. Henny, US Geological Survey, Corvallis, OR, R. Norstrom, Canadian Wildlife Service, Ottawa, ON. Osprey (*Pandion haliaetus*) eggs were collected from 1991 to 1997 at nests (N=121) upstream and downstream of bleached kraft pulp mills and at reference sites in the Fraser and Columbia River drainage systems. Samples were analyzed for polychlorinated dibenzo-p-dioxins (PCDDs), dibenzofurans (PCDFs), biphenyls (PCBs) and organochlorine pesticides. Mean concentrations of 2,3,7,8-TCDD were significantly higher in eggs collected in 1991 at downstream compared to upstream nests near pulp mills at Kamloops and Castlegar, British Columbia. There were no significant temporal trends in 2,3,7,8-TCDD, -TCDF or other measured compounds at nests monitored between 1991 and 1994 downstream of the Castlegar pulp mill, despite changes in bleaching technology (CLO2 substitution). However, by 1997 concentrations of 2,3,7,8-TCDD and -TCDF were significantly lower than previous years in nests sampled downstream at both Castlegar and Kamloops. An unusual pattern of higher chlorinated PCDDs and PCDFs was found in many of the osprey eggs, and both OCDD and 1234678-HpCDD concentrations were positively correlated with pentachlorophenol concentrations. Use of chlorophenolic wood preservatives by lumber processors was considered the main source of higher chlorinated PCDD/Fs throughout the systems. Concentrations of DDE and most other organochlorine pesticides showed high variability among individual eggs within study areas, but no significant differences in concentration among study areas. Mean PCB concentrations varied significantly among areas, with greater concentrations found in eggs from the Columbia compared to the Fraser basin, attributed to the extensive development of hydroelectric generation and associated industries on the former.

PTA101 Using PCB Congener Patterns to Identify PCB Sources. Beltman, D.*, Cacela, D., Lipton, J., Hagler Bailly Services, Boulder, CO. Multivariate statistical techniques were used to quantitatively and qualitatively characterize PCB congener patterns in sediment and biota samples in an attempt to identify the parent aroclors that were the sources of the PCBs observed in the samples. The samples were collected at increasing distance from known Aroclor 1242 point sources. The purpose of the analysis was to determine the extent to which congener patterns in samples from downgradient of the known Aroclor 1242 point sources were consistent with weathered Aroclor 1242, with other weathered aroclor patterns from other sources, or with a combination of aroclors. The analysis included consideration of PCB environmental chemistry, including dechlorination, volatilization, dissolution, and adsorption to sediment. The results showed that most of the samples contained PCBs in a pattern most consistent with weathered Aroclor 1242, indicating known point sources along the river as the dominant PCB sources to the river and bay. As distance from the point sources increased, the apparent contribution of aroclors other than 1242 increased (taking weathering into account). Several different multivariate techniques were used, and the efficacy of the different techniques for characterizing PCB congener patterns was compared.

PTA102 Regional assessment of organochlorine fish tissue contamination in northeastern U.S. lakes. R. Yeardley, Jr.*, SoBran Inc., Environmental, Cincinnati, OH; J. Lazorchak, S. Cormier and M.S. Berigan, U.S. EPA, Cincinnati, OH. A regional assessment of northeastern U.S. lakes indicated that, in many lakes, levels of organochlorine fish tissue contamination were in excess of values determined to be of potential health concern for wildlife and human consumers. The Environmental Monitoring and Assessment Program (EMAP)-Surface Waters conducted a survey of northeastern U. S. lakes during 1993-1994 and analyzed whole fish composite samples for organochlorine contaminants, including total PCBs, total DDT, total chlordane, dieldrin and other compounds. Distributions of the contaminants determined from this survey were evaluated to estimate the proportions of lakes in the northeastern region which exceeded human screening and wildlife values. Human screening values were obtained from the USEPA Office of Water and wildlife values were calculated based upon the USEPA Great Lakes Water Quality Initiative (GLWQI). Calculations indicated that approximately 99% of lakes contained fish with PCBs exceeding a human screening value of 0.01 µg/g and 90% of lakes contained fish with PCBs exceeding a mammalian wildlife value of 0.02 µg/g. Further, approximately 8% of lakes contained fish with DDT levels in excess of a human screening value of 0.3 µg/g and 48% had fish exceeding the mammalian wildlife value of

0.04 μg/g for the same contaminant. The usefulness of the EMAP survey sampling design, which allows regional assessment without a complete resource inventory, was practically demonstrated in this regional assessment of a class of contaminants.

PTA103 Distribution of Organochlorine Pesticides in Sediments and Benthic Organisms from Korean Coastal Areas. Lee, K.T.*, Seoul National University, Seoul, Korea; Tanabe, S., Ehime University, Matsuyama, Japan; Koh, C.H., Seoul National University, Seoul, Korea. The residues of organochlorine pesticides (HCHs, CHLs, and DDTs) were determined in 62 sediments and 5 invertebrates from Kyeonggi Bay and nearby areas along the West coast of Korea. HCHs were uniformly distributed at most sites studied whereas CHLs and DDTs showed a distinctive gradient of contamination between inner and outer sites of Incheon North Harbor, where these levels were one or two orders of magnitude higher than in the other sites. The distribution of CHLs and DDTs was correlated with TOC (total organic carbon) contents in sediments, while HCH residue levels were independent of TOC. The predominant organochlorine pesticide in sediments was β-HCH, *trans*-chlordane, and *p,p'*-DDD, whereas the most persistent compounds in marine benthic organisms were β-HCH among HCH compounds, *trans*-chlordane among CHL compounds, and *p,p'*-DDE among DDT compounds. The BSAFs (biota-sediment accumulation factors) of organochlorine contaminants in this study implied that β- and γ-HCH, *trans*-chlordane, *cis*-chlordane, *cis*-nonachlor, *p,p'*-DDD, and *p,p'*-DDE may be bioaccumulated from sediment.

PTA104 Estimating Dietary Exposure to Chemicals in Terrestrial Wildlife. Moore, M.L.*, Mackay, C.E., Mellott, R.S., Ginn, T.C., and Booth, P.N., Exponent, Bellevue, WA; Medved, J.B., General Motors Corporation, Detroit, MI. Risk to terrestrial wildlife from chemical exposure typically is evaluated by comparing species-specific estimates of dietary exposure to toxicity thresholds for selected ecological receptors. In many assessments, chemical concentrations in food items are estimated from chemical concentrations in soil and other environmental media using bioaccumulation factors (BAFs). Despite the broad use of BAFs in ecological risk assessment, the uncertainty associated with their use has not been rigorously evaluated. This paper evaluates the accuracy of published BAFs for predicting chemical concentrations in mice, earthworms, plants, and insects from soil concentrations using empirical data collected as part of an ecological risk assessment for a site in Michigan. Soils at the site contained elevated levels of barium, cadmium, chromium, copper, lead, vanadium, zinc, and polychlorinated biphenyls (PCBs). Ecological risks associated with exposure to these compounds are evaluated in four ecological receptor species: the northern short-tailed shrew (*Blarina brevicauda*), the white-footed mouse (*Peromyscus leucopus*), the American robin (*Turdus migratorius*), and the eastern screech owl (*Otus asio*). Species- and chemical-specific dietary exposures are estimated using a standard food-web model that incorporates either measured or BAF-predicted chemical concentrations in food items. BAF-based concentration estimates in food items are substantially higher than measured concentrations for certain food items and comparison of empirically and BAF-derived dietary exposure estimates indicates that the use of BAFs from the scientific literature may substantially overestimate dietary exposure.

PTA105 Application of the model BIOSCREEN to a TCE-contaminated sand aquifer in West Tennessee. Boston, A.N.*, Tennessee State University, Nashville, TN; Williams, S.D., and Byl, T.D., U.S. Geological Survey, Nashville, Tennessee. BIOSCREEN is a modeling system, based on the Domenico analytical solute transport model, designed to make predictions about the natural attenuation of dissolved phase hydrocarbons in ground water. BIOSCREEN can simulate advection, dispersion, adsorption, and anaerobic and aerobic reactions. BIOSCREEN also can approximate changes in the horizontal and vertical extent of a contaminant plume and can estimate how long the plume will persist in ground water. To make predictions about the transport of trichloroethylene (TCE) contamination in a West Tennessee sand aquifer, three input parameters of the model will be modified: (1) adsorption, (2) rates of biodegradation, and (3) dispersion. Parameter modifications are based on field and laboratory data from the site. Preliminary laboratory data indicate that adsorption may play a critical role in the natural attenuation of TCE in the sand aquifer. Batch-type experiments conducted using TCE concentrations similar to those found in the field showed that 50 percent of the TCE in the dissolved phase may be sorbed by aquifer material. Other field and laboratory data appear to indicate that biological degradation does not play a significant role in decreasing TCE concentrations in the aquifer at this time. Dissolved oxygen concentrations in ground-water samples (measured at 0.5 to 1.6 milligrams per liter) were too high to support reductive dechlorination and too low to support co-metabolism. Low concentrations of potential electron donors also may limit biological degradation of TCE in the aquifer.

PTA106 Role of Dissolved Organic Matter in Fenton Degradation of Hydrophobic Pollutants. Tarr, M.A.*, Lindsey, M.E., University of New Orleans, New Orleans, LA. Dissolved organic matter (DOM) can affect Fenton degradations in several ways. First, DOM can complex iron, altering its reaction with hydrogen peroxide and changing the formation rate of hydroxyl radical. Second, DOM can act as a natural scavenger and remove hydroxyl radical from the system, making it unavailable to react with pollutants. Third, DOM is known to bind hydrophobic pollutants, thereby altering the pollutant's microenvironment. Such microenvironmental changes may alter the pollutant's rate constant for reaction with hydroxyl radical, may alter the local concentration of hydroxyl radical relative to the bulk solution, and may result in different degradation products. We have developed chemical probe methods to measure the production and scavenging rate of hydroxyl radical in the presence of DOM. In addition, we have developed methods of producing steady state hydroxyl radical concentrations during Fenton oxidation. As a result, pseudo first order kinetics for pollutant degradation have been observed. These simplified kinetics have enabled the determination of pollutant-hydroxyl radical rate constants, and allow probing the effects of binding on degradation rate constants.

PTA108 Chemical Lability of Covalent Bonds Between Reductive Degradation Products of TNT and Peat. Thorn, K.A., USGS, Arvada, CO; Thorne, P.G., CRREL, Hanover, NH. One component of the chemistry underlying the bioremediation of TNT contaminated soils is the covalent binding of the monoamino- and diamino- reductive degradation products of TNT to soil organic matter. Because covalent binding to soil organic matter may be considered a way of detoxifying the amines, there is a need to understand the types of bonds formed, and their chemical stability under environmental conditions. Studies are therefore being conducted using ^{15}N NMR to determine how 4ADNT, 2ADNT, 2,4DANT, and 2,6DANT form covalent bonds with model quinone and lignin compounds, soil humic acid, and peat, in the presence and absence of enzyme and metal catalysts. Initial studies on the chemical lability of the conjugates are focused on the susceptibility to hydrolysis of the covalent bonds formed between the IHSS Pahokee peat and the mono- and diamines. In the absence of catalysts, all four amino compounds undergo nucleophilic addition reactions with carbonyl groups in the peat and become incorporated in the form of aminohydroquinone, aminoquinone, amide, and heterocyclic nitrogens. The diamines are more reactive than the monoamines, and condense with the peat to form imine bonds as well. Birnessite as catalyst increases both the total incorporation of the amines and the total proportion of condensation products as imines. A combination of base and acid hydrolysis released the amines bonded to the peat via aminohydroquinone, aminoquinone, amide, and imine linkages. Amines incorporated into the peat in the form of heterocyclic condensation products are stable to hydrolysis.

PTA109 Weathering-Induced Changes in Dissolved Petroleum Hydrocarbon Chemistry: Implications for Using WAF Derived Toxicity Bioassay Results. Haddad, R.I.*, Holder, J.L., DeMartino, L., Nedoff, J., ENTRIX, Inc., Walnut Creek, CA. Dissolved petroleum hydrocarbons (DPH) derived from a free phase product plume in groundwater can migrate to surface water where they pose a potential risk to the environment. Bioassays are often used to evaluate the potential toxicity associated with DPH. At least two different approaches can be followed in conducting these bioassays. The first approach uses petroleum hydrocarbons (PH) from the free phase plume in the preparation of a water accommodated fraction (WAF). Bioassays are conducted with the WAF to create a dose-response curve (DRC) which is then compared to DPH concentrations measured in surface water. The second approach uses bioassays conducted with site specific surface media. The benefits to the first approach include the ability to use the DRC and surface water DPH concentrations to derive toxicity information without the need for many site-media bioassays. This assumes that dilution is the only environmental process affecting the DPH chemistry. The benefit of the second approach is that there are no inherent assumptions regarding the DPH chemistry in the bioassay results. However, bioassays are required at each location. This study used high resolution gas chromatography and mass spectrometry to test the hypothesis that changes in DPH chemistry downgradient from a free phase plume are controlled solely by dilution. Comparison of DPH fingerprints from the free phase PH and from DPH samples collected along a downgradient transect clearly show this hypothesis to be incorrect. The data indicate that other processes (e.g., adsorption, biodegradation, etc.) have a measurable effect on the chemistry of the DPH. These results suggest care must be exercised in the field when using the first approach and that a detailed understanding of the specific environmental processes affecting the DPH chemistry is important in developing bioassay strategies.

PTA110 Prevention of Metal Diffusion from Contaminated Sediments Using *In situ* Caps Containing Coal Combustion By-products: Evaluation Using Redox Gel Probes. Edenborn, H.M.*, U.S. DOE, FETC, Pittsburgh, PA; Brickett, L.A., U.S. DOE, FETC, Pittsburgh, PA. One method used to keep toxic compounds in sediments from being released into the overlying water is *in situ* capping, which seals off contaminated sediments under one or more layers of soil, rock, or synthetic materials. In this study, we formulated several capping mixtures using neutral and alkaline coal combustion by-products, including various additives, such as lime, magnesium peroxide, and activated carbon. These mixtures were tested in a sludge pond receiving drainage from a flooded underground coal mine. Dissolved iron diffuses from the sediments in this pond, resulting in a relatively constant outflow concentration of ca. 10 mg/l. Various capping mixtures were added to core tubes placed in the pond sediment. The materials were allowed to settle within the tubes, then were maintained in constant contact with the overlying pond water. After incubation of these cores for 2 months, redox gel probes containing MnO_2 and FeS were pushed into sediments within and adjacent to the core tubes and were left for one week. After this time, the cores were collected, extruded, sectioned, and analyzed for porewater metal concentrations. Redox gel probes were examined in the field, and then sectioned and acid-digested for total metal content. Chemical analyses of the porewaters showed that all alkaline caps effectively retained iron in the sediment over the study period, whereas concentration gradients of conservative ions were quickly re-established across the caps. Redox gel probes containing MnO_2 provided a rapid assessment of the location of migrating redox boundaries, whereas probes containing FeS were only sensitive indicators of the depth of dissolved oxygen penetration into sediments.

PTA111 Transformation of Tetrachloroethene (PCE) at the Surface of Dithionite-Reduced Clay Minerals and Iron Rich Minerals Bryan Heath*, The University of Georgia, Athens, GA; Valentine Nzengung, The University of Georgia, Athens GA; Gary Mills, Savannah River Ecology Laboratory, Aiken, SC Mineral surfaces play an important role in the transformation of anthropogenic organic chemicals in the environment. Iron-bearing minerals are among the dominant constituents of many soils, sediments and aquifer materials. In fact, iron is the most abundant terminal electron acceptor in anoxic subsurface environments. Consequently, reducing Fe(III) in clays and aquifer materials to Fe(II) to create a passive and *in-situ* reactive zone could potentially be an effective approach for the remediation of chlorinated organic compounds, such as tetrachloroethene (PCE). The advantages of using dithionite reactive barriers are: (1) the natural occurring iron and other redox metals are utilized instead of constructing trenches packed with hundreds to thousands of tons of iron, (2) the reactive zone can be regenerated easily, (3) injecting dithionite into an aquifer is technically less challenging, (4) the approach can be more easily adapted to plume migration, and (5) the engineering costs are less than those of constructed zero-valent iron barriers. In this study, anaerobic batch studies were conducted to determine the effectiveness of dithionite-reduced clay minerals, iron rich minerals and aquifer materials to degrade PCE. The objectives were: (1) to determine the reaction rates of various mineral surfaces, (2) identify the factors that influence the reaction kinetics and (3) identify the reaction products and pathways. Five clay minerals (Na-montmorillonite, ferroginuous smectite, kaolinite, illite and vermiculite), four iron rich minerals (goethite, magnetite, hematite and siderite) and three aquifer materials (from Winton-Salem, North Carolina, Chamblee, Georgia and Tobacco road Formation at the Savannah River Site) were tested. Each of the aquifer materials were characterized using x-ray diffraction. Of the different clay minerals, iron rich minerals and aquifer materials tested, iron oxides exhibited the fastest degradation rates. The rate and extent of degradation of PCE by clay minerals was NOT dependant on the amount of iron present, but on its location in the clay mineral structure (i.e., *cis-* versus *trans*-position). The aquifer material degradation rate was dependant on the type and amount of clay minerals and iron rich minerals. The degradation products identified so far include: trichloroethene, dichloroethene, acetylene and ethene. The presence of the degradation products indicates that consecutive reductive dehalogenation is the dominant PCE transformation process.

PTA112 Dechlorination of Chloromethanes in Iron and Palladium-Iron Bimetallic Surface in Aqueous System. Wan, C., Chen, Y.H., and Wei, R.* Department of Chemistry, Cleveland State University, Cleveland, Ohio 44115. Iron granules (<10 μ) Fe^0-H_2O) and palladium treated granules (Pd/Fe°-H_2O) in contact with water have been tested as a potential means to dechlorinate chloromethanes ($CC1_4$, $CHC1_3$, CH_2C1_2) that are commonly generated by the teaching chemistry laboratories. Dechlorination of a rate of $CC1_4$ assumed pseudo-first order with respect to iron mass. As compared to the dechlorination rate of $CC1_4$ for the Fe°-H_2O enhanced the dechlorination of $CC1_4$ by a factor of about seven. The oxygen in the aqueous solution inhibited the dechlorination of $CC1_4$ by both Fe°-H_2O and Pd/Fe°-H_2O approximately one-half. From the consideration of remediating the solvent wastes, both systems therefore appear suitable for treating the $CC1_4$ and possibly $CHC1_3$ but the potential toxic effect of palladium remains a concern. Reactivity of three chloromethanes toward the Fe°-H_2O was vastly different. Dechlorination rate of $CC1_4$ and the $CHC1_3$ were systematically examined on both Fe°-H_2O and Pd/Fe°-H_2O. Based on the data, it appears that $CC1_4$ is successively dechlorinated to form CH_4 via the formation of partially dechlorinated intermediates, and the proton transfer to each of the intermediates then produced corresponding chloromethanes: $CHC1_3$, CH_2C1_2, and CH_3C1. Further, the CH_4 formation, independent of the proton transferring reactions, appears to be selectively stimulated by the palladium (Pd/Fe°-H_2O).

PTA113 Reductive Dechlorination of Polychlorinated Dioxins by Humic Materials Under Reducing Conditions. Fu, Q.*, Adriaens, P., The University of Michigan, Ann Arbor, MI. Experiments were conducted to investigate whether natural organic matter such as humic acid materials can facilitate the dechlorination of polychlorinated dioxins (PCDDs) under reducing conditions and to find correlation between humic acid structure characteristics with the kinetics of dechlorination of PCDDs. Reductive dechlorination of 1,2,3,4,6,7,9-heptachlorodibenzo-*p*-dioxin (heptaCDD) was carried out with one humic acid (AHA) and one model fulvic acid (PMA) in buffered medium with titanium citrate (III) as bulk electron donor. Dechlorination of heptaCDD with both AHA and PMA were observed. A ln(C/Co) vs time plot indicated a pseudo-first-order rate of disappearance of heptaCDD. The rate of disappearance is nearly doubled with AHA, and increased by a factor of 1.7 with PMA, as compared to the rate in the presence of titanium citrate (III) alone. Multilinear regression of the pseudo-first-order rate constants with various parameters characteristic of AHA and PMA was carried out to quantitatively correlate the rate constant with those parameters. The rate is highly correlated with the phenolic acidity. Up to five chlorine atoms were removed in all treatments, however, the individual congeners produced within each isomer group in each treatment were different. The major dechlorination products formed under the experimental conditions are hexa- and pentachlorinated congeners, accounting for more than 90% of all the lesser chlorinated products formed. The bottle neck appears to be dechlorination of pentaCDD to tetraCDD. The higher amount of lesser chlorinated products formed with AHA than those with PMA is consistent with the larger pseudo-first-order rate constant observed with AHA. The results of this investigation with humic materials contribute to our knowledge on the fate and transformations of PCDDs in the environment. The abiotic processes involving humic materials may greatly affect the rate as well as the reaction pathways of reductive dechlorination of highly chlorinated dioxins.

PTA113A

PTA114 Modeling Field Level Pesticide Drift onto Individual Ponds Using Remote Sensing and GIS Technologies. C. M. Holmes* and S. A. Kay, Compliance Services International, Tacoma WA; M. E. J. Waller and K. Z. Travis, Zeneca Agrochemicals, Berkshire, UK. This study utilized field level information to estimate potential drift deposition onto aquatic habitats using satellite imagery and a Geographic Information System (GIS). Field level information was generated from satellite imagery in which cotton was separated from other agriculture, and static water bodies were identified. Drift deposition for each water body pixel was calculated based on the drift contributed by each individual upwind cotton pixel. Each water pixel was examined from 8 different wind directions. Individual cotton pixels were classified as having ground application, aerial application or a combined ground/aerial application, each with their own associated drift potential. Drift deposition for all pixels in the water body were combined to compute the total deposition for the pond. Drift values were based on the AgDRIFT model developed by the Spray Drift Task Force. As an additional analysis, the classified land cover was used to identify tree pixels found between cotton and water. In these cases, the drift values from cotton pixels were modified to allow for the estimated drift reduction factor contributed by the trees. The results of this study are highly localized spray drift calculations based on the actual spatial distribution of cotton and trees around individual water bodies. These results show total estimated drift deposition onto a pond based on application method, as well as identify varying drift deposition across the pond, drift results for each wind direction, and the drift reducing effect of trees found between cotton and water. Because this analysis was performed at an individual pond level, the results of this type of field level analysis can lead to more realistic exposure estimates and probabilistic modeling.

PTA115 Model Uncertainty: A Tool for Use in Model Selection. Snowling, S.D.*; Kramer, McMaster University, Hamilton, ON, Canada. Evaluation of uncertainty in environmental modelling is fundamental to the assessment of risk and, the use of modelling in risk management. The inherent uncertainty of a model is hypothesized to be in part a property of the complexity of the model structure and assumptions made in its development. By choosing a model of appropriate complexity, an environmental manager can control the amount of relative uncertainty associated with the results. The question is: how much uncertainty is associated with a given model complexity? Using both model error (how well a model can simulate reality) and model sensitivity (how sensitive the model output is to the quality of input) to define model uncertainty, the uncertainty-complexity relationship has been developed for a series of groundwater transport models. Multiple models of differing complexity were used to simulate tracer plume movement at the CFB Borden site in Borden, Ont. For each model, the model error and model sensitivity were assessed, and results showed that improvements in model error are offset by associated increases in model sensitivity. Specifically, going to more complex models (for this particular system and model set) does not decrease modelling uncertainty, since the inherent sensitivity of the more complex models outweighs the relatively minor improvements in model error. This type of test provides information that can be used by an environmental manager for the purposes of model selection, with the objective of balancing the need for better simulations of real systems with the cost of developing and providing data for increasingly complex models. The decision of how much sensitivity and error is acceptable is made by the manager based upon the objective of the risk assessment, and modelling resources (e.g. data, money) available.

PTA116 The Use Remote Sensing Imagery to Characterize Wildlife Habitat on a Regional Scale. La Tier, A. J. and Macdonald, B.; EVS Environment Consultants, Seattle, WA and North Vancouver, BC. One of the goals of conducting an ecological risk assessment is to quantify exposure of sensitive species to chemical contamination to predict potential risk. Ideally, site-specific information is collected to characterize habitat as part of the process. However, financial resources are often lacking to conduct the necessary fieldwork, or the site is large enough to warrant a more sophisticated approach. In order to conduct more "ecologically relevant" risk assessments, we propose the use of remote sensing imagery to identify plant communities and habitat types. Habitat information can be linked to spatial contamination contours in a geographic information system to determine exposure point concentrations for ecological receptor species. Remote sensing data are available at various levels of resolution, and will be used as available to compare the ability of these sensors to resolve and delineate target habitats. In our presentation we will provide an example of the process for developing a habitat characterization for a selected study site. We will present the process for selecting ecological receptor species within the limitations of the various levels of remote sensing data available. We will then outline the habitat requirements of those species and present the approach for retrieving, utilizing, and analyzing the remote sensing data to quantify receptor species exposure. The purpose of using various remote sensing sources is to provide a guide to resolution vs. habitat recognition, and not as a definitive study on a selected habitat. To fulfill this purpose, satellite and aerial data will be used to compare data content and cartographic accuracies needed to map species habitat.

PTA117 Dose-response Curves and Patch Arrangement: Implications for Field Extrapolations and Risk Assessment. Lenart, L.A*, Landis, W.G., Institute of Environmental Toxicology and Chemistry, Huxley College, Western Washington University, Bellingham, WA. A critical issue in environmental toxicology and risk assessment is the extrapolation from laboratory to the field. Typical benchmarks in toxicology/risk assessment only provide point estimates such as EC10, LC05, or NOEC to compare to environmental concentrations. Although this is a convenient number to use, the midpoint or other defined point of a sigmoidal curve does not define the curve. The shape of the curve may determine population and landscape impacts. Three different dose-response curves, each with the same midpoint, were used in the metapopulation model developed at our research facility to determine changes in impacts to population dynamics. Patch arrangements used in these series of simulations represented increasingly fragmented landscapes. We found that although connected patches were not dosed, they were indirectly affected by the dynamics of the dosed patch. The largest effect was seen in the linear or least connected arrangement of patches. Different population growth patterns were also seen by changing the dose-response curve with the shallower curves having the greatest effect. Impacts were also defined by the configuration of the patches. In some cases the patch farthest from the dosed patch showed effects long after the disappearance of the environmental stressor. Dramatic differences also occur depending on whether the dosed patch is a source or a sink. These results demonstrate that extrapolation of a dose-response curve to ecological effects is dependent on specific knowledge about the initial conditions of the populations and parameters that define the spatial dynamics. Only site-specific risk assessments will contain this level of information for such an extrapolation.

PTA118 Regularities observed in species sensitivity distributions. De Zwart, D.*: Van de Meent, D., National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands. For a large number of both organic and inorganic toxicants, log-logistic species sensitivity distributions are fitted to acute and chronic data obtained from the AQUIRE database (US-EPA, Duluth, USA). The log-logistic sensitivity model is characterized by two parameters only: (i) α (alpha) which is the average of the observed log transformed $L(E)C_{50}$ or NOEC values over a variety of test species, and (ii) β (beta) which is proportional to the standard deviation of the log transformed toxicity values. A regression of acute and chronic alpha values for a large number of chemicals reveals that the average acute toxicity is a factor of 10 higher in concentration than the average chronic toxicity. The same type of analysis on the acute and chronic beta values demonstrates that the factorial difference between the sensitivities of the most and least sensitive species tested is equal for both acute and chronic tests. Provided that sufficient species are tested, the magnitude of beta values proves to be related to the toxic mode of action of the compounds considered.

PTA119 Potentially Affected Fraction as an Indicator of Toxic Stress; Application to Aquatic and Terrestrial Ecosystems in The Netherlands. Van de Meent, D., Klepper, O., De Zwart, D.*, National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands. Interspecies variations of No Observed Effect Concentrations are used to relate field concentrations of toxic substances to stress on ecosystems. Given a concentration in water or soil the proportion of species for which the NOEC is exceeded is calculated. This proportion is called the Potentially Affected Fraction (PAF), and is used as an indicator of toxic stress on ecosystems. PAF is calculated for individual substances; combi-PAF is calculated for mixtures of substances. PAF-values for individual substances are used for chemicals on the basis of the toxic stress put on ecosystems. Combi-PAF values for mixtures are used for comparing polluted sites. Both PAF and combi-PAF are used for following trends over time. Toxic stress on terrestrial and aquatic ecosystems in The Netherlands was assessed by PAF calculation for heavy metals, pesticides and other priority pollutants). The toxic potency of surface waters was also measured experimentally. The results suggest that both aquatic and terrestrial ecosystems are severely stressed by exposure to man-added metals and pesticides. Narcotic organic chemicals appear to play a lesser role.

PTA120 Combining toxicants in a single indicator for toxic stress. De Zwart, D.*; Van de Meent, D.; Aldenberg, T., National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands; Hamers, T., Wageningen Agricultural University, Dept. of Toxicology, Wageningen, The Netherlands. For ecotoxicological risk assessment purposes, the environmental concentration of a single compound can be expressed as the fraction of species which is exposed to a bioavailable concentration above its NOEC: the Potentially Affected Fraction of species (PAF). In general, specific pollution situations are characterized by the presence of a multitude of pollutants. In order to estimate the combined toxic risk at a particular site, assumptions have to be made about the interactions between chemicals and about the types of species to be affected. Two clear-cut options are available. (i) If several substances in the environmental cocktail do have exactly the same toxic mode of action, their concentrations in terms of Toxic Units (TU) can be added (concentration additivity) prior to calculating their PAF. The concentration-additive substances are considered to demonstrate their effects in the same collection of most sensitive species (species dependency). The factorial difference between the most and least sensitive species has proven to be equal for compounds with the same mode of action. (ii) Other (groups of) substances, with a different toxic modes of action are considered to exert their effect on a different set of most vulnerable species (effect additivity). For the situation of pure effect additivity the combined PAF can be calculated by applying species independency rules. Especially pure species independency may not be a fully realistic assumption, which may lead to an over-estimation of the combined risk.

PTA121 Ecological effects of exceeding No-Observed Effect Concentrations (NOECs) of persistent toxicants on soil organisms. Klepper, O.; Traas, T.P*.; Schouten, A.J.; De Zwart, D., National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands; Korthals, G.W., Wageningen Agricultural University, Dept. of Nematology, Wageningen, The Netherlands. Estimating the effect of toxic substances on ecosystems generally has to rely on frequency distributions of species sensitivity based on a limited variety of laboratory tests. Information on the sensitivity of local species is normally lacking. Toxic stress can be expressed as the fraction of generic species exposed above their NOEC: the Potentially Affected Fraction of species (PAF). This paper describes a model of the soil foodweb and the effect of toxic stress by persistent pollutants. The model predicts that in the absence of competition individual species will disappear from the foodweb at toxic concentrations 3-5 times their NOEC. With competition present, species affected by toxic stress are replaced by less sensitive species. This has a two-fold effect: (i) species disappear from the foodweb at a lower concentration because of competitiveness occurs well before absolute extinction; (ii) the replacement of extinct species implies that the effect on total biomass and diversity can only be detected at PAF levels near 100%. Model predictions are in good agreement with effects observed in nematode communities exposed to field soils experimentally contaminated with copper and zinc. The model illustrates why overall measures of ecosystem functioning (total biomass, production, diversity) are only affected by high levels of toxic stress. The apparent robustness of ecosystem functioning masks a considerable erosion of ecosystem structure in terms of sensitive species which is properly reflected by the PAF.

PTA122 Conducting Regional Risk Assessments of Air Pollutant Impacts on Vegetation using Uncertainty Modeling and Spatial Simulation. Woodbury, Peter B*.: Laurence, John A. Boyce Thompson Institute for Plant Research, Ithaca, NY. In order for decision-makers to rationally address complex environmental problems, a methodology is needed for making science-based decisions with incomplete information. This methodology must organize what is known, what can be concluded by credible extrapolation, and how sensitive the conclusions are to what is still unknown. We have developed such a methodology and used it to model the effects of tropospheric ozone on soybean yield for each county in the USA where soybeans are grown. A geographic information system (GIS) was created to integrate regional data such as soybean yield, likelihood of drought, and estimated cumulative ozone exposure. Spatially continuous regional data sets were created by interpolation when required and uncertainties in these data sets were estimated. The GIS was linked to a Latin Hypercube simulation model in order to produce probabilistic estimates of yield loss due to ozone. This model permitted us to quantify how the effect of ozone on yield in each county was influenced by uncertainties in critical factors such as ozone exposure, extrapolating from experimental data to farmer's fields, interactions between drought and ozone, and estimation of dose from ozone exposure. The model predicted that current ozone concentrations have a greater than 50% chance of causing a substantial decrease in yield throughout much of the USA. Regional variation in the effect of ozone was observed, due primarily to variation in ozone exposure and soybean yield. We show how this methodology can improve assessments of ecological risk by identifying regional patterns in the influence of critical factors on the response of vegetation to ozone and identifying where uncertainty in model predictions is the greatest.

PTA123 A Revised Approach to Applying Uncertainty Factors in the Ecological Effects Assessment of New Substances in Canada. Bonnell, M.*, Bonnell Environmental Consulting, Ottawa, ON, Canada; Atkinson, A., Environment Canada, Hull, QC; Hammond, G., Environment Canada, Hull QC; Postlethwaite, E., Environment Canada, QC. The ecological effects assessment of new substances in Canada requires the use of uncertainty (safety) factors to estimate toxicity threshold concentrations of chemicals or polymers notified under the New Substances Regulations of the Canadian Environmental Protection Act (CEPA). A study was conducted to "update" the traditional USEPA-based approach currently used to assess new substances in Canada in order to provide a more transparent, data-derived method to determining uncertainty factors. The revised method estimates a residual level of uncertainty based on the calculation of component *certainty factors* that account for pivotal study quality, inter- and intra-specific variation, acute to chronic extrapolation, severe effects to no effects extrapolation and laboratory to field extrapolation. The results of five new substance case studies, which compared the revised approach with the USEPA-based approach, and conclusions and recommendations of the study are also presented.

PTA124 Geographically-referenced Environmental Assessment Tool for European Rivers: GREAT-ER. Validation by Monitoring in UK Catchments. Holt, M. (ECETOC, Brussels, BE), Fox, K. K.* (Unilever, Port Sunlight, UK), Daniel, M., (Environment Agency Laboratory Service, Leeds, UK), Morris, G., (Environment Agency, Leeds, UK), McEvoy, J., (Environment Agency, Thames, UK), and Buckland, H. (Yorkshire Water, Bradford, UK). The monitoring work which has been carried out in four catchments in the United Kingdom to validate GREAT-ER, a Geographic Information System (GIS) model being developed to predict the concentrations of consumer product chemicals in European rivers, is described. Linear alkylbenzene sulphonate, LAS, which undergoes both adsorption and biodegradation in sewage treatment works (STWs) and in rivers, and boron, a conservative chemical, are the chemicals chosen for model validation. The sampling programme, which comprised over 2500 samples from regular Environment Agency (EA) river monitoring sites and STW effluents, gives a record of LAS and boron concentrations and of water quality determinands obtained simultaneously, over a 1-2 year period. Measured distributions for LAS, boron, and water quality parameters obtained at effluent and river sampling sites within the catchments will be presented, and are compared with simple model predictions of the river water concentrations, including the GREAT-ER model. The results are also compared with consumer usage data, and show that up to 50% of LAS removal takes place in Yorkshire sewers. The validation results for these pilot catchments show that GREAT-ER will deliver a considerable improvement over current European methods for predicting the concentrations of consumer chemicals, when the data necessary to populate the model for the whole of Europe become available.

PTA125 Screening Soils for Potential Groundwater Vulnerability Using a Modified SEEPAGE Model with STATSGO Soils Data in a Geographic Information System. S.A. Kay*, C.M. Holmes, and R. Lemus, Compliance Services International, Tacoma, WA; J.P. Carbone, Rohm & Haas Company, Spring House, PA. As the source of drinking water to more than half of the US population and providing almost a third of the water used for irrigation, groundwater issues have risen to the forefront of environmental debates in the past 15 years. The EPA has estimated that agricultural activities are the most serious and widespread cause of water quality problems from nonpoint sources in the US today, with the most frequently identified nonpoint sources considered to be fertilizer and pesticide applications and septic systems. Embracing the philosophy that protection of groundwater might be best accomplished by prevention of contamination, a number of screening tools have been developed to identify areas of most serious pollution potential. This study utilized a nationwide soils database (STATSGO) and a modified version of the SEEPAGE model implemented in a Geographic Information System (GIS) to quantitatively rank each soil's vulnerability to ground water contamination. Combined with agricultural suitability information, it is possible to examine leaching vulnerability in relation to specific crops. This poster illustrates the methodology used to generate the vulnerability data set and the role of the GIS in the analysis. The results that can be obtained from the data set are presented in both tabular and graphical (map) format.

PTA126 Assessment of the Potential Refractory Toxicity of Organic Water-soluble Chemicals with the PC-based Computer Program AQUATOX™. Diehl, R.A.*, and Moore, S.B., Burlington Research, Inc., Burlington, NC. Review of production process, non-commercial housekeeping/cleaning, and water treatment chemicals used by industries and in wastewater treatment operations in an important component of a toxicity reduction evaluation in assessing potential sources of refractory toxicity in final effluent. To facilitate the evaluation of a chemical's potential for contributing refractory toxicants or its affect on activated sludge treatment systems, a PC-based computer program has been developed to estimate environmental impact using recognized OECD indices: 28-day biodegradability, 48-h acute LC50, and C-h sludge inhibition EC50. Using data based on product actives, a weighted ration of endpoints is employed to calculate a numerical rating that designates a chemical's potential environmental impact as "Low", "Moderate", "High", or "Very High". With this guidance, chemical usage can be optimized to maximize the use of "Low" and "Moderate" impact chemicals to the greatest extent possible.

PTA127 A Validation Trial of LEACHP Using Independent Field Lysimeters. Carbone, J.P.*, Rohm and Haas Co. Spring House, PA. The pesticide component of the Leaching Estimation and Chemistry Model (LEACHP) (Version 3.1) was used to evaluate the leaching potential of an experimental herbicide and its relevant metabolites. The Rohm and Haas Co. recently completed a field study where the herbicide was applied to a series of 1-meter deep lysimeters with a cover crop. The use of multiple field lysimeters offered a unique opportunity for the conduct of a calibrated and tested or "validated" LEACHP modeling analysis. Because of the multiple lysimeters installed, LEACHP could be calibrated against the measured data of a single lysimeter and then the calibrated model could be validated against several independent lysimeters. Comparisons of measured and model estimated hydrologic parameters, specifically recharge and soil pore volume were highly correlated. Bromide movement while correlated in terms of mass flux and concentration was not correlated with regard to event timing. The reason for the event timing discrepancy is speculative but may be a consequence of model limitations, measured data variability or the soil model employed in conjunction with the highly sandy nature of the soil modeled. Calibration of the transformation/degradation rate parameters enabled validated simulations of lysimeters to generate mass flux; soil pore water and soil core concentrations of the parent and relevant metabolites that were typically within a factor of 2 – 5 of measured data. Event timing was moderately correlated. The validation trial illustrates an ideal approach for the use of environmental fate and transport modeling for the analysis of the leaching potential of a crop protection chemical.

PTA128 Analyzing the Unsteady State Fate of DDTs in Lake Maggiore (Italy): Data vs. models. Di Guardo, A.*, Terzaghi, G., Calamari, D., Department of Structural and Functional Biology, University of Milan, Varese VA, Italy. Lake Maggiore is a lake situated in North-west Italy, on the borders of Piedmont, Lombardy and Switzerland, and is the second largest Italian lake, after Garda. In 1996 DDT was found in fish at much higher levels than the previous average background concentrations (in the order of hundreds of micrograms per kilogram). Also the most recent sediment concentrations varied between 0.1 and 1 mg/kg d.w. In order to understand the fate of DDT and metabolites (o,p'-DDT, p,p'-DDT, o,p'-DDE, p,p'-DDE, o,p'-DDD, p,p'-DDD) in the lake and the surrounding area a monitoring activity of biota at different trophic level (phyto- and zooplankton, benthos, forage and piscivorous fish), of water (lake and rain), suspended sediment and bottom sediment was initiated. The initial results of forage fish (*Coregonus spp.*) concentrations show lower concentrations (around tens of micrograms) than those initially obtained. A lake model with a food-web submodel was developed in order to organize existing data and predict future behavior. The first results of the monitoring activity and model simulation are presented in an attempt of understanding the unsteady-state trends. Results suggest that most of the uncertainty in the model simulation is related to the difficulty of obtaining accurate values for key intermedia parameters such as sediment deposition/resuspension velocities.

PTA129 Use of Process Modeling Software to Support Process Development, Process Fit Analysis, and Environmental Analysis; Case Study: Pharmaceutical Process. Hettenbach, K.*, Pfizer, Inc., Groton, CT; Ericson, J., Pfizer, Inc., Groton, CT. Process modeling software is an efficient and cost effective tool for performing process development tasks and for scaling up processes into full scale production. Process modeling simulation tools speed process development, manages change faster, and provide a material and environmental assessment analysis, as process are scaled from the Process R&D/Pilot Plant stage (*Conceptual* modeling) to Full Scale Production (*Process Fit* modeling). Process modeling is a viable tool for integrating the chemist task with that of the process engineer. Process modeling may also be used as an effective tool to assess process optimization during development. Batch Design Kit is a process modeling tool designed for the pharmaceutical batch industry and is used to illustrate the above features.

PTA130 Application of Monte Carlo Simulation in Ecological Risk Assessment: Moving from Conservative Assumptions to Realistic Estimates of Exposure and the Potential for Adverse Effects. Kubitz, J., and Friant, S.L. ENTRIX, Inc. A tiered process is commonly used to assess the potential for unacceptable risks to ecological receptors at facilities that have been affected by chemical releases. In the first tier, very conservative (i.e. worst-case) assumptions are typically used to develop screening criteria. By comparing site data to the screening criteria, the risk assessor can determine which chemicals will not pose an unacceptable risk to ecological species under worst-case conditions. In the second tier, site-specific data and assumptions are used to calculate a Hazard Quotient. It is not uncommon to have incomplete site-specific information, so conservative assumptions are used to address the uncertainties that result. A combination of several conservative assumptions can produce an unrealistic exposure scenario. If the second tier of an ecological risk assessment concludes that unacceptable risks could occur, and that expensive remediation is needed, it can be cost-effective to use a third tier, probabilistic risk assessment (PRA), to investigate the risks associated with realistic exposure scenarios. The results of a PRA are a distribution of the probability of an unacceptable risk, and a sensitivity analysis that describes which factors are contributing the most to the unacceptable risk. A case study will be used to illustrate the differences in interpreting the results of Hazard Quotient and Probabili\stic Risk Assessments for making risk management decisions.

PTA131 Ecological Risk Analysis Incorporating Interspecies Interactions in a Japanese Lake. Miyamoto, K.*[1,4], Matsunaga, T.[1], Iida, M.[1], Naito, W.[2], Masunaga, S.[2,4], Nakanishi, J.[2,4], Bartell, S. M.[3], [1]National Institute of Materials and Chemical Research, Tsukuba, Japan, [2]Yokohama National University, Yokohama, Japan, [3]*SENES* Oak Ridge Inc., Oak Ridge, TN., [4]CREST, Japan Science and Technology Corporation. The objective of this study is to develop a method for evaluating the potential risk posed by chemicals to aquatic ecosystems in Japan. Population dynamics of 40 species from 11 categories of trophic levels in Lake Biwa was modeled, including predator-prey interactions and struggle for existence, under time-varying environmental conditions, using the Comprehensive Aquatic Simulation Model. To determine individual growth, physiological processes such as photosynthesis (for primary producers), consumption (for consumers), loss to predation, non-predatory death, and respiration were formulated and changes in the rate of these processes due to toxic chemicals were estimated from laboratory toxicity data. The risks posed by chemicals were calculated as probabilities of the decrease in biomass using Monte Carlo simulation. The simulated biomass of phytoplankton, zooplankton, and fish populations without toxic stress was compared with the biomass observed in Lake Biwa. Major results and conclusions are as follows. Seasonal changes in the simulated biomass without toxic stress agreed relatively well with those from actual observations. Chemicals such as pentachlorophenol, which are more sensitive to upper trophic levels than to lower ones, were estimated to directly decrease the biomass of the upper levels. Some chemicals such as DDT had the possibility to reduce even insensitive species because of reduction in food when their preys were sensitive. Sensitivity analysis indicated that the photosynthesis rate was the key parameter that affected the biomass of producers. This work was supported by Core Research for Evolutional Science and Technology of Japan Science and Technology Corporation.

PTA132 Pathogenesis of Ulcerative Dermatitis and Myositis in Coastal North Carolina Fish. Law, J.M.* and Marquart, M., North Carolina State University, Raleigh, NC. Recent massive fish kills in eastern North Carolina, often associated with "sores" or ulcerative lesions in certain fish, have brought *Pfiesteria piscicida* to the forefront of public attention. This tiny dinoflagellate, recently dubbed the "cell from hell" by the popular media, has been blamed for most of these kills. However, the actual cause of these fish die-offs has yet to be proven. In particular, the prevalence and pathogenesis of the ulcerative dermatitis and myositis, usually centered on the anal pore of Atlantic menhaden (*Brevoortia tyrannus*), is poorly understood. The purpose of the present study was to determine the cellular and tissue-specific stages in lesion development, along with the temporal and spatial distributions of ulcerative dermatitis and myositis in stratified random samples of fish from the Neuse, Tar/Pamlico, and New River estuaries of North Carolina. In order to determine the extent of ischemic injury in relation to ulcer development, we employed a battery of markers of ischemia-reperfusion injury and endothelial cell activation. Our results suggest that these lesions develop in an "inside out" fashion due to sublethal hypoxia. That is, focal ischemic necrosis of the deep dermis and adjacent musculature appears to be the primary lesion, followed by secondary invasion of fungal and bacterial organisms and subsequent focally severe inflammation.

PTA133 Monitoring for Toxics in Waters Demonstrating Toxic *Pfiesteria* Episodes: Exploring a Possible Contributing Factor. Kroll, R.B.* and Eskin, R.A. Maryland Department of the Environment, Baltimore, MD. In the late summer and fall 1997, a series of fishkill episodes occurred in several water bodies in Maryland's lower eastern shore, including the Pocomoke River. The fishkills occurred in the absence of other commonly encountered causes (e.g., very low dissolved oxygen, chemical spills) in waters also demonstrating significant incidents of fish lesions (deep, bleeding, and often circular sores) considered "typical" of those associated with "*Pfiesteria*-like" organisms. In response to these episodes, the State undertook a large study to determine possible causes for the fishkills and lesions. Although scanning electron microscopy and fish toxicity bioassays conducted by other investigators subsequently confirmed the presence of toxic life stages of several dinoflagellates, including *Pfiesteria piscicida*, the State continued investigating other possible contributing factors. Current scientific knowledge suggests the most likely contributing factors are the natural physical characteristics of the River (especially the broad shallow mouth and deep channel in the upper river), and certain chemical characteristics, both natural (e.g., deeply colored water limiting light penetration) and anthropogenic (e.g., nutrient enrichment). However, high concentrations and/or chronic exposure to certain chemical contaminants are known to cause lesions and kill fish. To explore toxics as a possible factor in the Pocomoke episodes, concentrations of chemical contaminants in water, sediment, and fish tissue were evaluated. Intensive monitoring for toxics in ambient waters following rain/runoff events was also conducted in fall 1997 and spring 1998. Data evaluated through early spring 1998 show no evidence that chemical contaminants (metals or pesticides) have contributed to the incidence of fish lesions or fishkills in the Pocomoke River. Monitoring continues.

PTA134 Use of a Reporter Gene Assay for Low Level Detection of *Pfiesteria piscicida* Toxins and its Potential Use in Evaluating Natural Water Samples. Deamer, N.J.[1]; Glasgow, H.B.[1]; Manning, J.P.[1]; Fairey, E.R.[2]; Edmunds, S.J.[2]; Moeller, P.R.[2]; Ramsdell, J.S.[2]; Burkholder, J.M.[1] [1] NCSU, Raleigh, NC[2] NOS, Charleston, SC. *Pfiesteria piscicida* is a toxic dinoflagellate that has been implicated as a causative agent in fish kills and disease events along the mid-Atlantic coast (from Delaware to Florida). *P. piscicida* is lethal to both finfish and shellfish at levels > 300 cells/ml and causes sublethal effects at 150 cells/ml. NOAA-NOS Charleston Laboratory has developed a GH4C1-A1 rat pituitary cell line that stably expresses a c-foc luciferase reporter gene, a known biomarker for several algal toxins. NOAA has developed this assay to assist in the toxin purification and detection. Initial studies indicate that partially purified *P. piscicida* toxin (P-Tox), as well as, straight laboratory toxic *P. piscicida* culture gave parallel luciferase induction concentration curves; wherein an increase in expression was observed over two log orders of concentration. Time course studies indicated that the water-soluble P-Tox induces the c-foc-luc approximately twice as fast as maitotoxin (the most toxic dinoflagellate toxin known to date). The mode of action for maitotoxin (i.e., directly on calcium channels) and P-Tox appears to be different. Nimodipine, a voltage dependent calcium channel antagonist, had no effect on P-Tox ability to induce c-fos-luc. The initial studies indicate that the reporter gene was sensitive/inducible to as little as 4-10 cells well^{-1}, which is equivalent to concentrations reported to be lethal to finfish (400-1000 cells ml^{-1}). This c-fos luciferase reporter gene assay is an important tool that may be suitable for evaluating natural estuarine waters. We are initiating field validation studies and will report these findings for the 1998 summer fish kill season in NC.

PTA134a Human Visual Function in the North Carolina Clinical Study on *Pfiesteria piscicida*. Hudnell, H.K., U.S. EPA, RTP, NC. The U.S. Environmental Protection Agency assisted the North Carolina Department of Health and Human Services in conducting a human-health study to investigate the potential for an association between fish kills in the NC estuary system and the risk for adverse human-health effects. Impetus for the study was recent evidence suggesting that the estuarine dinoflagellate, *Pfiesteria piscicida*, may release a toxin(s) which kills fish and adversely affects human health. This report describes one component of the study in which visual system function was assessed. Study participants worked primarily in estuaries inhabited by *P. piscicida* or in off-shore waters thought not to contain *P. piscicida*. The potentially exposed estuary (N=22) and unexposed offshore (N=20) cohorts were well matched for age, gender, and education, but less well matched for occupation. Visual acuity did not differ between the cohorts, but visual contrast sensitivity (VCS), an indicator of visual pattern-detection ability for stimuli of various sizes, was significantly reduced in the estuary cohort relative to the offshore cohort. A further analysis which excluded participants having a history predictive of neuropsychological impairment also showed significantly reduced VCS in the estuary cohort (N=14) relative to the offshore cohort (N=10). Additional analyses indicated that differences between the cohorts in age, education, smoking, alcohol consumption, and total time spent on any water did not account for the difference in VCS. Finally, an analysis which excluded members of the estuary cohort who may not have had direct contact with an active fish kill, as well as offshore participants who may have had direct contact, also indicated that VCS was significantly lower in the estuary (N=17) than the offshore (N=17) cohort. The profile of VCS deficit across stimulus sizes resembled that seen in organic-solvent exposed workers, but an assessment of solvent and other neurotoxicant exposures did not indicate differences between the cohorts. These results suggest that factor(s) associated with the NC estuaries, including the possibility of exposure to *P. piscicida* toxin(s) at active fish kills, may impair visual system function.

PTA135 Evaluation of the Developmental Toxicity of Amlodipine besylate. Orisakwe, O.E.,* Chilaka, K.C., Afonne, O.J., Obi, E., Nnamdi Azikiwe University, Nnewi Campus, Anambra State.Amlodipine besylate was evaluated for its potential to cause embroyonal/fetal toxicity and teratogenicity in pregnant mice. Amlodipine was administered through drinking fluid at dose levels 5 and 20 ug/ml on days I through 21 of gestation. Fetuses were examined on gestational day 21. There was significant ($p<0.05$) decrease of the relative weight of heart, liver, uterus, pancreas and vagina in the 20 ug/ml amlodipine while the 5ug/ml amlodipine produced a significant ($P<0.05$) decrease in the relative weight of the pancreas and vagina. Amlodipine showed a significant ($P<0.05$) dose dependent increase in maternal body weight and total white cell count. There was no growth retardation of embryo.

PTA136 Embryo and Larval Toxicity of Alkylphenol Exposure in *Fundulus heteroclitus*. Kelly, S.A.* and Di Giulio, R.T., Duke University, Durham, NC. Alkylphenol polyethoxylates comprise a major class of nonionic surfactants that are degraded to more persistent alkylphenols, such as nonylphenol (NP) and octylphenol (OP). Both alkylphenols have been identified in the aquatic environment and both are considered estrogenic. In previous work, we reported that NP and 4-*tert*-octylphenol (4-*t*OP) were embryotoxic to *Fundulus heteroclitus*, an estuarine teleost. We also reported that tamoxifen, an estrogen receptor antagonist, could partially alleviate the sublethal toxicity of NP. Here we report that tamoxifen can rescue embryos in lethal concentrations of NP and 4-*t*OP. This rescue effect is more pronounced with NP: (1) embryos can survive higher molar concentrations of NP than 4-*t*OP when tamoxifen is present, and (2) in comparing the toxicity of each alkylphenol at the same molar concentration, the degree of rescue is greater for NP. However, while the embryos coexposed to tamoxifen survive in lethal alkylphenol concentrations, sublethal abnormalities are still evident. Further studies evaluating hatch success and subsequent larval behavior of embryos exposed to NP and 4-*t*OP have shown that these endpoints are also adversely affected. This has been observed in embryos exhibiting relatively mild sublethal abnormalities. Tamoxifen does not appear to alleviate effects on hatch success and larval behavior. Notably, coexposure of NP and 4-*t*OP produces toxicity that appears greater than additive in both embryonic and larval endpoints. Interestingly, larval exposure to the alkylphenols suggests that NP and 4-*t*OP are lethal to larvae at concentrations at least an order of magnitude less than those that are embryo-lethal. Additional studies are evaluating perturbations in apoptosis as a mechanism of the observed developmental toxicity.

PTA137 Variation of Light and Temperature Regimes and Resulting Effects on Medaka (*Oryzias latipes*) Reproduction. Koger, C.S.*, Teh, S.J., Hinton, D.E. University of California, Davis, Davis, CA. Changes in fecundity, fertility, and brood interval have been associated with changes in temperature and photoperiod. To determine their impact in quantitative terms and to determine whether alterations in embryo or larval viability follow variations with these cues, a light tight environmental water bath with controllable natural light and temperature was developed. Forty groups, two breeding pairs per group, were subjected to varying photoperiod and temperature regimes. Each group produced 20±3 embryos daily at 16L/8D and 25 ÌC. Histologic analysis of gonads revealed abundant mature ova and spermatozoa with no oocyte atresia. Cessation of embryo production followed a change to 8L/16D (25ÌC) and a severe decline in production was observed after a change to 15 ÌC (16L/8D). Subsequent histologic analysis showed no mature ova and severe oocyte atresia at 8L/16D (25ÌC). However, mature ova and mild oocyte atresia were seen at 15 ÌC (16L/8D). At both low photoperiod and temperature regimes, mature spermatozoa were observed. Embryo and larval viability, along with brood interval, were not affected by a change in photoperiod. Temperature change had no effect on embryo or larval viability, but caused an increase in time between broods. Results indicated that photoperiod rather than temperature profoundly affected medaka reproduction. Decreased temperature reduced but did not arrest fertility. Photoperiod affected fecundity. None of the changes in reproduction was permanent. After changing conditions back to 16L/8D and 25ÌC, embryo production resumed at control levels, although histologic data indicated a residual effect on oocyte atresia. These defined conditions are being used to determine *in vivo* effects of endocrine disrupting compounds on reproduction in this teleost model.

PTA138 Effect of Octylphenol on Serum Vitellogenin, Reproductive Capacity, and Offspring of Male Medaka Fish. Gronen, S., and Brouwer , M*. University of Southern Mississippi. Institute of Marine Sciences. Exposure of male fish to (xeno)estrogens results in elevated levels of vitellogenin (VTG) in their blood. To examine whether VTG can be linked to reproductive impairment, we exposed adult male medaka (50 fish/treatment and control; 4 treatments and 2 replicates/treatment and control) to 4-*tert*-octylphenol (OP) for a three week period. After 21 days, OP exposure was terminated and blood was collected from eight fish from each aquarium for measurement of VTG by Western blot analysis. On day 22, 30 female fish were added to each aquarium. Eggs were harvested each morning for 11 days and counted and microscopically assessed for fertilization. At the end of the egg collection period, ten males from each treatment and control were bled for determination of VTG. The progeny generation was initiated with embryos harvested from treatment and controls during the reproductive phase of the study. Histological examination of the testes of OP-exposed fish showed the presence of oocytes in some of the fish. There were significant correlations ($p<0.01$) between OP exposure concentrations and VTG in serum of male fish. VTG at end of reproductive study had decreased between 70-90% compared to VTG measured immediately after exposure. In addition, there were significant correlations ($p<0.01$) between VTG in serum of fish at the start and end of the reproductive study and (1) Percentage of fertilized eggs; (2) Percentage embryo survival and (3) Incidence of abnormal embryo development (loss of vascular integrity, bilateral microphtalmia, unilateral anophtalmia, and small yolks). It appears that vitellogenin in male fish can be used as a predictive indicator of impaired reproduction and that OP has both estrogenic and genotoxic properties.

PTA139 Effects of water-borne 4-nonylphenol on Atlantic salmon (*Salmo salar*) smolts. Brown, S.B.*, National Water Research Institute, Burlington, ON; Haya, K., Burridge, L. St. Andrew's Biological Station, St. Andrew's, NB; Taylor, E.O., Arsenault, J.T., Fairchild, W.L., Gulf Fisheries Centre, Moncton, NB. Our recent evaluation of relationships between historical applications of Matacil® 1.8D to forested areas of Atlantic Canada to control damage from spruce budworm (*Choristoneura fumiferana*) and catch data for Atlantic salmon populations suggest possible impacts. Matacil® 1.8D contains the carbamate insecticide aminocarb with 4-nonylphenol (4-NP) as a diluent. After spraying, reported aminocarb concentrations in water samples were well below reported lethal thresholds for Atlantic salmon. However, the estimated 4-NP concentrations fell within a range where estrogenic effects in fish might be anticipated. The spray program was coincident with final stages of smolt development in salmon. When historical salmon catch data were evaluated considering effects on survival at the smolt stage, there was a significant negative relationship between the returns of large Atlantic salmon and the proportion of individual tributaries sprayed within the Restigouche River drainage basin in 1977. There was also a broader event of unusually heavy salmon smolt mortality in 1977. This smolt mortality event contains a significant relationship indicating that where Matacil® 1.8D spraying occurred, the smolt mortality increased. For 19 spray events occuring in 14 rivers, 53 % of the lowest salmon catches between 1973 and 1990 coincided with times when Matacil® 1.8D had been sprayed. To begin experimental investigations about the hypothesis that the 4-NP in the Matacil® 1.8D formulation may represent the suspected causal agent, we exposed Atlantic salmon to enviromentally relevant, pulse doses of waterborne 4-NP during the latter stages of smoltification and report on their capability to withstand sea water challange.

PTA140 Effect of DDT and Estradiol on Gonadal Development and Liver Pathology in Juvenile Male Summer Flounder *(Paralichthys dentatus).* Zaroogian, G.*, Gardner, G., Borsay, D.,Haebler, R., Gutjahr-Gobell, R., Mills, L., U.S. EPA, Narragansett, R.I. Xenobiotic compounds that disrupt endocrine function target the same organs as native hormones and affect the development and sexual maturation in vertebrates. Among the best known xenobiotics that bind with the estrogen receptor is o,p'-DDT. The intent of this study was to compare histopathologically the effect of o,p'-DDT and 17β-estradiol (E2) on gonadal development and liver condition in sexually immature male summer flounder *(Paralichthys dentatus).* Two year old juvenile male summer flounder were injected into a dorsal sinus with 15, 30 or 60 mg/kg of o,p'-DDT or 0.1, 1.0 or 10 mg/kg E2 incorporated in coconut oil. A second identical injection was administered two weeks later. Fish were sampled at 4, 6, and 8 wks after the initial injection and observed histopathologically. In fish treated with 30 or 60 mg/kg o,p'-DDT, spermatogenesis regressed significantly $p<0.001$) when compared to controls. E2 treatment of 1.0 or 10 mg/kg elicited altered gonadal development similar to that observed with o,p'-DDT but in fewer fish. No consistent pattern of toxic injury to the liver and biliary tract was observed in o,p'-DDT treated fish. Diffuse liver injury, necrosis in random clusters of cells, loss of normal array of hepatocytes and intracytoplasmic pigmentation occurred after 4 wks in all fish treated with 10 mg/kg E2 and after 6 wks in 50% of the fish treated with 1.0 mg/kg E2. Submassive necrosis developed in 45% of fish treated with 10 mg/kg E2 after 4 wks.

PTA141 Evaluation of Vittelogenin in Japanese Medaka, *Oryzias latipes* **Following Oral Exposure in a Multigeneration Study.** P. J. Patyna* and K. R. Cooper. Joint Graduate Program in Toxicology. Rutgers-The State University of New Jersey, Piscataway, NJ. Vittelogenin (VTG) production in male fish serves as a biomarker for exposure to estrogenic compounds. In previous studies 17 β-estradiol fed for 129 days at 50, 500, and 5000 ppb resulted in 100% feminization of males. 17β-estradiol treated fish, fed at 50ppb for 10 days were used as positive controls for the production of VTG. Western blot analysis was performed using primary monoclonal mouse anti-striped bass antibodies. Analysis of whole liver cytosolic protein in 17 β-estradiol treated fish revealed two subunits of VTG at 170 kDa and 130 kDa. No lower molecular weight protein cross reactivity was detected. VTG levels were evaluated in Japanese medaka chronically exposed for 255 days to diisononyl phthalate (DINP) and diisodecyl phthalate (DIDP) (20 μg/g) in a multigeneration feeding study. VTG was not detected in male F_0 generation fish from Untreated, DINP, DIDP and Acetone control groups. Female F_0 Untreated, DINP, DIDP fish showed similar VTG levels at 170 kDa, no 130 kDa band was observed. VTG was not detected in F_0 Acetone treated females. No VTG was detected in any F_1 male treatment group. F_1 female DINP, DIDP, and Acetone control groups showed a marked increase in VTG when compared to the Untreated F_1 females. Treatment with DINP and DIDP did not result in the detection of VTG in male fish in either F_0 or F_1 generations. There was a solvent related increase in female VTG levels in the F_1 generation. These biochemical findings are in agreement with egg fecundity, viability and growth data (Supported by ES22330, ES29782, and NJAES01126).

PTA143 Investigations of Potential Endocrine Disruption and Sexual Dimorphism in Nestling Tree Swallows (*Tachycineta bicolor*) with a Range of PCB Body Burdens. Yorks, A.L.*, Rattner, B.A., Melancon, M.J., USGS Patuxent Wildlife Research Center, Laurel, MD; Bakst, M.R., USDA, Beltsville Agricultural Research Center, Beltsville, MD. Polychlorinated biphenyls (PCBs) elicit endocrine disruptive effects in many species, including birds. Tree swallows (*Tachycineta bicolor*) were studied at eight sites, located in Maryland, Pennsylvania, and New York, with a range of PCB contamination to determine effects on gender and gonadal development of nestling offspring. Blood samples were collected from nestlings and genetic sex was determined by polymerase chain reaction amplification of sex chromatin in nucleated red blood cells. Gonads were excised and fixed for subsequent gross and histologic examination. PCB analyses of twelve-day old nestlings indicated that residue concentrations varied considerably among the eight sites. Of the 145 nestlings examined anatomically, the phenotypic sex ratio was 53% female and 47% male. No intersexes were observed. Histological observations revealed some variation such as numbers of spermatogonia and stages of follicular development among individuals. Genotypic evaluation of the 145 nestlings revealed complete concordance with phenotypic observations. Although there were significant differences in PCB exposure among study sites, there was no evidence of abnormal gonadal development or anatomical gender alteration in nestling Tree swallows.

PTA144 Ovotestes in male common tern embryos (*Sterna hirundo*) do not persist to the prefledgling stage. Hart, C.A.*, Hahn, M.E., Nisbet, I.C.T., Woods Hole Oceanographic Institution, Woods Hole, MA; Kennedy, S.W., Environment Canada, Hull, QC. Common and roseate terns breed on Bird Island, Massachusetts near a Superfund site highly contaminated with PCBs. The possibility of contaminant related endocrine disruption in these birds was first suggested by skewed sex ratios and female-female pairings among endangered roseate terns. This prompted sampling of common tern embryos as a surrogate species. In 1993 and 1994, 70-90% of pipping male common tern embryos sampled showed the presence of ovarian cortical tissue in their testes (ovotestes). To examine the possibility that the presence of ovotestes could affect reproductive capabilities in common terns, the persistence of ovotestes was examined. Common tern prefledglings (approximately 21 days old) were collected from Bird Island, and their gonads were examined histologically. In addition, contaminants were measured in a subset of previously collected eggs from the same nests as a measure of embryonic contaminant exposure. None of the 19 common tern prefledgling gonads examined showed the presence of ovotesticular tissue. Some gonadal irregularities were observed, including small nodules of testicular tissue within the epithelial capsule of the testes, but these were judged not likely to affect testicular function. There was no relationship between any observed irregularities and levels of contaminants present in the matched eggs. Although 70-90% of pipping common tern embryos showed development of ovotestes, none of the prefledgling common terns had ovotestes, thus suggesting that ovotestes in common terns have fully regressed by approximately 21 days. It is unlikely that the presence of ovotestes would result directly in abnormal testicular function. (Supported by NOAA Sea Grant and an EPA Graduate Fellowship).

PTA145 Extent and Magnitude of Reported Effects of Polychlorinated Biphenyls, Dioxins, and Furans on a Colonial Piscivorous Bird, the Double-Crested Cormorant (*Phalacrocorax auritus*). Schmeising, L.M.*, Exponent, Boulder, CO; Wallin, J.M., Iannuzzi, T.J., and Ludwig, D.F., Exponent, Landover, MD. A review of field and laboratory studies was conducted to evaluate the reported extent and magnitude of toxicological effects from polychlorinated biphenyls (PCBs), and polychlorinated dibenzo-*p*-dioxins and polychlorinated dibenzofurans (PCDD/Fs) to the double-crested cormorant (*Phalacrocorax auritus*). The objectives of this investigation were to: 1) determine the reported effects of these compounds on a widely occurring species that is at particular risk of exposure to these and other chemical compounds in waterways throughout the world, and 2) evaluate the available dose-response and determine trends and similarities in the reported effects thresholds. Data from available studies on waterways throughout the world were compiled and evaluated for comparability of toxicological endpoints, study design, quality, and reported effects thresholds. Comparable data were tabulated and subjected to statistical analyses to develop the appropriate distribution form for the data set. Commonly reported effects include reduced hatching and fledging success, chick wasting syndrome, and edema, embryo mortality, and malformations in embryos and chicks. Congener-specific information for PCB and PCDD/F mixtures were also evaluated to determine the incremental contribution of these compounds to the reported effects based on commonly used toxicity equivalence schemes. The results suggest that the reported toxicity of these compounds must be considered with respect to the overall toxicological activity of the group of chemicals known as Ah-receptor-active compounds, as well as to confounding variables such as habitat factors that effect population fluctuations and survivorship of these birds.

PTA146 Brain Asymmetry, Behavior and Related Effects in Domestic Hatchling Chicks Following Developmental TCDD Exposure. DeWitt, J. C.*, Meyer, E. B. and Henshel, D. S., Indiana University, Bloomington, IN. Ongoing studies in our laboratory have revealed that brain asymmetry is detectable in experimental bird models following 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) exposure and follows trends similar to those seen in birds environmentally exposed to mixtures containing TCDD. Similarly, behavioral studies performed in our laboratory have revealed that dose-dependent behavioral differences correlate with dose-dependent differences in brain asymmetry. Further analysis of the exposed hatchling chicks was explored to determine relationships between brain asymmetry, behavior and responses such as time to hatching and sex ratios. Fertile, domestic chicken eggs were injected into the air sac prior to incubation with 2, 20 or 200 pg TCDD/g egg diluted in sunflower oil. Controls received the oil alone or no injection. Eggs were incubated and upon hatching, behavior of chicks was analyzed with several simple tests that assessed locomotion, perception and reflexes. Chicks were sacrificed within 24 hours of hatching. Brains were quickly removed after sacrifice and both the body and the brain were immediately formalin-fixed. Once fixed, dorsal, ventral and lateral measurements (12 total) were made on each half of the forebrain and the tecta and organs were necropsied and weighed. The resulting behavioral, brain asymmetry and dose-response curves of other responses were compared. The dorsal and lateral forebrain measurements correlated best with a reflexive test (righting reflex) and the amount of time between pipping and hatching. The lateral forebrain measurement also correlated best with two locomotor tests (activity and running time) and the crown-rump length. Finally, the dorsal tectal measurements correlated best with a locomotor test (gait analysis) and the M:F sex ratio. These results indicate that similar trends exist between organ effects, brain asymmetry and behavior.

PTA147 Monoclonal antibodies that cross react with vitellogenins from quail, terns and barn swallows. Chow, M.,* Kroll, K., Sapp, L., and Denslow, N.D. University of Florida, Gainesville, FL., Shutt, L., Lorenzen, A., Canadian Wildlife Services, Deem, S.L., White Oak Conservation Center, and Gross, T., USGS-BRD, Gainesville FL. Vitellogenin (Vtg) expression in males is a sensitive biomarker for exposure to environmental estrogens. We have developed monoclonal antibodies to monitor endocrine disruption that results from exposure to estrogen mimics in birds. Vtg expression was induced in male quail by slow release estradiol implants consisting of 1.25 mg b-estradiol/bird and in male terns by injection of 1 mg/Kg body weight per day for 7 days of b-estradiol. Vtg was purified from tern and quail plasma samples by ion-exchange chromatography on MonoQ using the BioCad Chromatography System. Egg yolk was purified from barn swallow eggs and a fraction composed mainly of a large protein, 120 kDa, corresponding to lipovitellin I, was used. A mixture of the two Vtg's and the lipovitellin I-rich fraction was injected into mice to generate antibodies. Hybridomas were made and tested for their cross-reactivities with the three different antigens. Using a direct ELISA screen with a mixture of the three antigens, we obtained a total of 31 antibodies. Of these 22 reacted with tern, 23 reacted with quail and 16 reacted with barn swallow samples. Of these 11 antibodies cross reacted with all three antigens, but only one mAb 3C3 cross reacts both by ELISA and western blot. We have purified and have begun to characterize mAb 3C3 for its specificity. Using mAb 3C3 we have developed and validated an ELISA assay for tern Vtg. We are now investigating the use of this antibody to measure Vtg induction in terns exposed to environmental estrogens. This reagent should add another instrument to the toolbox for testing endocrine disruption in birds.

PTA148 Sublethal Effects of Endosulfan on the Grass Shrimp *Palaemonetespugio*. Wirth, E.F.*, Fulton, M.H., Scott, G.I. National Ocean Service, Charleston, SC, USA. The grass shrimp, *Palaemonetes pugio*, is a commonly used toxicity test organism and a vital species within estuarine systems. Historically contaminated areas have been shown to have reduced grass shrimp populations, but the cause of these declines has not been well characterized. Reproduction in crustaceans has been shown to be affected by laboratory spiked sediments (pesticides and PAHs) as well as contaminated field collected sediments. The goal of this study was to investigate the potential for contaminant-induced reproductive declines in the grass shrimp using endosulfan (an organochlorine pesticide) as a model toxicant. Acute lethal and sublethal toxicity tests were performed and physiological methods were developed to assess the relationship between reduced offspring production and endosulfan exposure. Protocols were developed to 1. quantify the crustacean juvenile hormone, methyl farnesoate in shrimp and 2. quantify an egg specific protein using an ELISA. These protocols will help to evaluate endosulfan effects on reproduction. Preliminary data suggest methyl farnesoate levels differ between males and females. Additionally, the ELISA used to quantify the egg protein is a relatively sensitive test and appears promising. Acute 96-h LC5O determinations indicated that males were nearly twice as sensitive to endosulfan as females (0.92 ug/L to 1.99 ug/L). Future research will focus on the effects of sublethal endosulfan exposure on hormone and protein levels as well as hatching success.

PTA149 The Effects of Contaminants in Fish on the Glochidialuccess and Juvenile Survival of Freshwater Mussels (Unionids). Kernaghan, N.J.*, Ruessler, D.S., and Gross T.S., USGS/BRD/Florida Caribbean Science Center, Gainesville, Florida. There are almost 300 species of unionid mussels in North America, of which approximately 70% (213 taxa) are considered endangered, threatened or of special concern. The introduction of contaminants into the aquatic environment has been identified as a possible cause for the decline of mussels and numerous studies have investigated the effects of selected contaminants on several species of freshwater mussels. However, to-date there have been no studies to determine the effects of contaminant body burdens in host fish which are parasitized by the glochidia of freshwater mussels. This study examines the effects of high body burdens of several different contaminants in fish on. Individual host fish were implanted with time-release pellets for DDE, Toxaphene, or Atrazine. Three fish were prepared at each concentration and control fish were implanted with placebo pellets. Glochidia were cultured using an *in vivo* (on fish) technique and transformation to juveniles took from 10 days to three or more weeks, depending on the species. Once transformed, juvenile mussels were transferred to moderately hard (hardness 80 - 100 mg/L) water and survival was monitored using a standard 9-day test protocol. Cessation of both activity and heartbeat was used as the measurement endpoint indicating death. Counts of live and dead were made daily and dead mussels removed. Results indicate that exposure to fish containing high body burdens of DDE during transformation significantly reduces the survival of juvenile unionid mussels. However, survival of juveniles transformed on fish containing high body burdens of Toxaphene did not appear to be adversely affected.

PTA150 PCB uptake in tissues and eggs of reproductively active oysters (*Crassostrea virginica*) after dietary exposure. F-L. E. Chu, L. A.Cruz-Rodriguez*, P. Soudant, and R. Hale, Virginia Institute of Marine Science, School of Marine Science, Gloucester Point, VA 23062. Reproductively active oysters were fed PCB-sorbed algal paste for 15 and 30 days and the uptake and distribution of PCBs in different tissue compartments (visceral mass, mantles, gills, and adductor muscles) and eggs were examined. PCB-sorbed algal paste was prepared by mixing PCBs (mixture of Aroclor 1242, 1254, and 1260) in acetone with algal paste (*Tetraselmis maculata* and *Thalassiosira weissflogii*; 5μg :1 g algal paste). The PCBs readily sorbed (99.9995%) to the algal paste. Oysters from Damariscotta River, Maine, were maintained in individual containers and fed daily with 0.2 g algal paste containing 0, 0.1, or 1.0 μg PCBs. After 15 and 30 days of exposure, oysters were spawned, eggs collected, and then tissues dissected. Preliminary analysis of eggs revealed that PCBs were accumulated by the oysters and transferred to the eggs. PCB content in the eggs of oysters exposed to 0.1 μg PCBs daily was 5.1 fg PCBs /egg after 30 days of exposure. PCBs in the eggs of oysters exposed to 1.0 μg PCBs daily ranged from 9.5 to 14.8 fg PCBs/egg at 15 days after exposure and was 57.8 fg PCBs/egg at 30 days after exposure. It is also noted that PCB exposure reduced the concentrations of two essential polyunsaturated fatty acids, eicosapentaenoic and arachidonic.

PTA151 Tributyltin Concentrations in Puget Sound and Narragansett Bay and the Current Health of Associated Populations of Gastropods. Birchenough, A.*, Evans, S.M., Dove Marine Laboratory, United Kingdom. In the United States, Puget Sound and Narragansett Bay are both centres of shipping activity. Mollusks living in them have therefore been subjected to TBT contamination for more than 25 years. The study will describe investigations of the current health of populations of gastropods abiding in both areas. These will be based on measurements of imposex, as biological indicators of tributyltin pollution, and the size structure of populations, as indicators of fecundity. We assessed population-level effects at (i) 'hot-spots' of contamination, such as marinas and dry docks; (ii) those on shores adjacent to major shipping lanes; and (iii) those on shores which are distant from shippin activity. We also investigated sites visited in the past by other authors to look at changes in these populations over time.

PTA152 Effects of Endocrine Active Chemicals on the Development of Sex Characteristics of *Daphnia magna*. Olmstead, A.W. and LeBlanc, G.A., North Carolina State University, Raleigh, NC. Daphnids have been used extensively to assess the effects of environmental chemicals on invertebrate reproduction. However, the susceptibility of sexual components of the life cycle of this organism to chemical exposure has been largely ignored. The goals of the present study were to: 1) develop routine and reliable methods for the production of male *Daphnia magna* in culture; 2) identify and characterize sex characteristics associated with both male and female daphnids; and 3) evaluate the effects of chemical exposure on the development of sex characteristics in this species. Decreasing the food (*Selenastrum capricornutum*) concentration in cultures by a half when the cultured daphnids were six days old stimulated male offspring production. Neonatal male and female daphnids were nearly indistinguishable and could be discerned only by the longer first antennae of the males. Male and female sex characteristics progressively developed during juvenile and early adult development. Anatomical sex differences included larger first antennae in males, morphological differences in the head capsule and carapace edge, setae on the carapace edge of males, and a prominent abdominal process in females. Females grew significantly more rapidly and attained a larger size as compared to males. Exposure of daphnids to the steroidal androgen androstenedione elicited changes in the development of sex characteristics suggestive of masculinization. Studies are underway to evaluate the effects of chemicals known to alter hormone metabolism and function on the development of sex characteristics.

PTA153 Population decline and organotin compounds in the imposex-induced ivory shell. Horiguchi,T.*, Shiraishi,H., Morita,M., National Institute for Environmental Studies, Tsukuba, Ibaraki, Japan; Hamada,F., Tottori Association of Propagation and Aquaculture, Tohaku-gun, Tottori, Japan; Kajikawa,A., Tottori Prefectural Fisheries Experimental Station, Tohaku- gun, Tottori, Japan; Shimizu,M., Nihon University, Fac. Biosciences, Fujisawa, Kanagawa, Japan. Total catch of the ivory shell, *Babylonia japonica* has remarkably decreased in Japan since the 1970s. Especially in Tottori Prefecture, the number of both eggs spawned by adults and juveniles artificially released into the sea, as well as its total catch, remarkably decreased since the 1980s. Recovery of total catch of the ivory shell has not been still observed although much effort has been continuously carried out to enhance the ivory shell stock in Tottori. The ivory shell is one of the Japanese gastropods which have been observed to be affected by imposex. Both penis and vas deferens were well-developed in the imposex individual. Oviduct blockage by vas deferens formation, however, was not observed. Results on the histo-pathological observation of gonads will be shown and discussed. Tissue concentrations of organotin compounds, such as butyltins and phenyltins, were determined by GC-FPD. Tributyltin (TBT) and triphenyltin (TPT) concentrations in tissue of the ivory shell were higher than those in other gastropods collected at the same area. Remarkable accumulation of TBT was observed in heart, ctenidium and osphradium, and high concentration of TPT was detected in ovary. Both TBT and TPT concentrations in ovary were positively correlated with penis length in the imposex individuals. It is possible that population decline was caused by reproductive failure accompanied with imposex induced by organotin compounds from antifouling paints in the ivory shell.

PTA154 A Community Survey of TBT Contamination in the North Sea using Imposex in Dogwhelks as a Biological Indicator of Pollution. Fletcher, H.*, Evans, S.M. Dove Marine Laboratory, United Kingdom. The North Sea is one of the busiest shipping areas in the world and, according to the *North Sea Quality Status Report 1995*, is severely contaminated by TBT. This paper presents the results of a 1998 survey of coastal areas of the Sea, using imposex in the dogwhelk *Nucella lapillus* as a biological indicator of TBT pollution. Assessments of the health of populations of *N. lapillus* are based on measures of abundance, numbers of juveniles (as indicators of fecundity), and imposex. These will be used to comment on the extent to which regulations introduced in the late 1980s, prohibiting the use of TBT-based paints on vessels less than 25m in length, have been effective in reducing contamination. This project was carried out by members of the local community, including high school students, undergraduates, postgraduates, schoolteachers, and members of NGOs, working in collaboration with both communities from other countries which border the North Sea and with scientists from the Dove Marine Laboratory. It is part of the Dove Marine Laboratory's community education programme which is entitled: *The North Sea: Our Joint Responsibility.*

PTA155 Development of a Ribonuclease Protection Assay for Vitellogenin mRNA in Fathead Minnows as a Screen for Estrogen Receptor Agonists. Korte, J.J.*, Kahl, M.D., Jensen, K.M., Pasha, M.S., and Ankley, G.T., U.S. EPA, Duluth, MN. The existence of vitellogenin in the plasma of male oviparous organisms has been established as evidence of exposure to chemical agonists of the estrogen receptor. A screening method for detection of these chemicals in fathead minnows (*Pimephales promelas*) is desirable because of the utility of this species as a model in short-term life cycle tests assessing reproductive and developmental toxicity. Because of the small size of this fish and the difficulty in obtaining tissue samples, the ribonuclease protection assay (RPA) was chosen as a test method because of its potential for greater sensitivity, specificity, and reproducibility than existing methods that focus on detection of a protein. To accomplish our objective, fish were injected with estradiol to induce the synthesis of vitellogenin mRNA. After extracting the mRNA and making cDNA by reverse transcriptase, a vitellogenin-specific segment of cDNA was amplified by polymerase chain reaction (PCR). For PCR, we used consensus primers that were designed based on multiple sequence alignments of known vitellogenin sequences from other species. After sequencing the amplified segment of cDNA, primers specific for fathead minnow vitellogenin cDNA were obtained and used to generate templates that could be used to synthesize antisense RNA probes for the RPA. Additional sequence information obtained by rapid amplification of cDNA ends (RACE) methods will be presented and compared with other vitellogenin sequences. We will compare the utility of the RPA method to methods based on detection of vitellogenin protein such as western blotting and ELISA.

PTA156 Gene Induction Fingerprints in Liver Tissue of Sheepshead Minnows Exposed to Estradiol, Diethylstilbestrol, and Ethynylestradiol. Bowman, C.J.*, Lee, H.S., Ferguson, R.J., and Denslow, N.D., University of Florida, Gainesville, FL and Hemmer, M.J., Webb, R., and Folmar, L.C. U.S. EPA, Gulf Breeze, FL. There is significant interest in defining critical endpoints that can be used to assess endocrine disruption in wildlife. While the goal is to assess physiological endpoints that may indicate altered growth, reproduction and development, these are generally difficult to quantify because of natural variability in populations. Since hormones act primarily by inducing genes in target tissues that account for the physiological changes observed, we have concentrated our efforts at that level. We are compiling a list of specific genes that are induced by estrogen or estrogen mimics in fish exposed to low concentrations in water. These studies are based on differential display PCR. We have exposed sheepshead minnows to a series of environmentally relevant doses of estrogen, DES and ethinyl estradiol in a flow through diluter system over a three week period. We have noted the induction of Vtg protein in the plasma as early as 24 hrs after the start of the treatment. We have prepared RNA from the livers and have used it to obtain gene induction fingerprints for these three known estrogenic compounds. The fingerprints match well with each other and with earlier experiments with pharmacological levels of estradiol administered by injection. This approach should enable us to catalog and compare effect observed with these compounds and may provide insight in determining some of the mechanistic effects of exposures to the various environmental contaminants.

PTA157 Effects of Steroids and Environmental Estrogens on Expression of Japanese Medaka Sex Steroid Hormone Receptor Genes. Colley, William C.* and Benson, William H., The University of Mississippi, University, MS. Fish are exposed to a diverse array of enviromental chemicals that are introduced into water supplies. A major cellular target for many environmental chemicals are the sex steroid hormone receptor proteins. We report the isolation of partial complimentary DNA clones for the androgen and progesterone receptors from the Japanese medaka (*Orizias latipes*) and the isolation of multiple complimentary DNA clones for a medaka estrogen receptor gene. The ER cDNA clones, isloated by Reverse Transcription PCR using oligonucleotide primers flanking the open reading frame, represent distinct messenger RNA isoforms derived from a single medaka ER gene via alternative splicing. The medaka ER mRNA isoforms are expressed in a tissue- and sex-dependent manner. The number of and the relative amounts of each isoform changes in response to a 24 hour exposure of fish to 17b-estradiol (350 picomoles per liter) as compared with unexposed fish. The medaka AR and PR cDNA's were cloned from male testis and female ovary, respectively, using RT-PCR with degenerate oligonucleotides based on conserved regions within vertebrate AR and PR genes. The partial medaka AR cDNA clone is 87% homologous to the human AR protein. This region of homlogy includes human AR amino acids 618 to 749. The partial medaka PR cDNA clone is only 77% homologous to the human progesterone receptor (from amino acids 833 to 909) but is 96% homologous to a reported cichlid (*Tilapia mossambica*) gluccocorticorticoid receptor. Both the medaka AR and PR genes are differentially expressed in male versus female liver and gonads. We are currently testing other steroids and several xenoestrogenic compounds for their effects on medaka ER, AR, and PR gene expression.

PTA158 Effect of Cadmium and Aroclor 1254 on Mcf-7 cells and Reproductive Toxicity in Female Deer Mice. Gentles,B.A., Alston, D.M., Smith, E.E., The Institute of Environmental and Human Health, Texas Tech University, Lubbock, TX. Finlay, M., McCoy, G., Benedict's College, Columbia SC. Toxic effects of cadmium and Aroclor 1254 (PCB), singly and combined were investigated by determining their effect on proliferation in MCF-7 cells, uteri and ovaries histology of deer mice (*Peromyscus maniculatus*). MCF-7 cells were exposed to Cd (1μM) singly and in combinations with PCB at concentrations of 1μM, 0.1μM, and 0.01μM for 48 or 72 hr. Cadmium at 1μM significantly increased cell proliferation as compared to controls, following 72 hr exposure. Aroclor 1254 significantly reduced proliferation at 1μM, however at the lower concentrations of 0.1μM and 0.01μM proliferation increased significantly, after 48 and 72 hr exposure, indicating that toxic response may be dose dependent. Combination of Cd and PCB 1254 at these concentrations significantly increased proliferation. Suppression of cell proliferation by 1μM PCB was markedly reduced in the presence of Cd (1μM). Asynchronous deer mice were treated with either 5 ppm PCB, 2 ppm Cd, 20 ppm Cd, or 2 ppm Cd/PCB and histology of the ovaries and uteri were assessed. Morphology of ovaries and uteri in all treatment groups except 20 ppm Cd appeared normal. Twenty ppm Cd induced a decrease in uterine lumen diameter and endometrial gland density. The glands were significantly smaller and cylindrical when compared to controls. Further research will be necessary to evaluate *in vitro* and *in vivo* toxicological effects of Cd and PCBs both singly and in combination.

PTA159 Estrogenicity of Selected Benzophenones in the *Saccharomyces cerevisiae*-based *lac*-Z Reporter Assay. Schultz, T.W.* and Sinks, G.D. College of Veterinary Medicine, The University of Tennessee, Knoxville, TN. A systematic investigation of the relationship between molecular structure, estrogenic activity (EC_{50}) and cytotoxicity (LC_{50}) of 16 selected benzophenones was conducted. The *Saccharomyces cerevisiae*-based *lac*-Z reporter assay was used. Estrogenicity was measured colormetrically as b-galactosidase activity. The present studies confirmed several structural features associated with estrogenicity. Benzophenone, the parent compound, as well as 2-hydroxybenzophenone were found non-estrogenic, however, if the polar moiety (i.e., hydroxyl group) is moved to the *para*-position, 4-hydroxybenzophenone, b-galactosidase activity is exhibited. Replacing the *para*-hydroxyl group with an amino group moiety decreased b-galactosidase activity. Replacement of the *para*-hydroxyl group with a methyl, chloro, methoxy, or nitro substituent eliminated activity. Moreover, comparisons of estrogenic potencies of polyhydroxylated and/or chlorinated benzophenones illustrate the importance of molecular symmetry to estrogenicity. A comparison of estrogenicity (EC_{50}) with cytotoxicity (LC_{50}) reveals that for those compounds that elicit b-galactosidase activity there is typically two to three orders of magnitude difference between EC_{50} and LC_{50} values.

PTA160 Effects of Cadmium and Aroclor 1254 on Protein Synthesis and Hsp90 in Deer Mice Testes. Burge, K.R.*, Alston, D.M., Smith, E.E., TIEHH, Texas Tech University, Lubbock,TX; Finlay, M., McCoy, G. Benedict's College, Columbia SC. In an effort to further understand the relationship between exposure to environmental contaminants and testicular responses, deer mice were exposed to one of five treatment groups: 2 ppm Cd, 20 ppm Cd, 5 ppm PCB, 2 ppm Cd/PCB and negative controls. Protein synthesis and hsp90 expression were analyzed using SDS-PAGE and western blot. All treatment groups induced synthesis of proteins with M_r of 66 kDA, which were absent in the control. An induction in protein synthesis with proteins bands with M_r of 99 kDA and 51 kDA occurred following 2 ppm Cd and 2 ppm Cd/PCB treatment. Aroclor 1254 (5ppm) singly and combined with cadmium, induced synthesis of proteins with M_r of 90 kDA and 29 kDA. Further characterization of the 90 kDA protein band using monoclonal antibodies demonstrated that the hsp90 protein was influenced by treatment. Densitometry reading indicated that 5 ppm PCB increased free hsp90, while 2 ppm Cd/PCB suppressed free hsp90. This data indicates that cadmium and PCB can influence testicular protein synthesis in deer mice, and is in agreement with histological indication of male reproductive toxicity from our laboratory.

PTA161 Evaluation of the Testicular Toxicity of Hexavalent chromium and effects of Zinc ii in Mice. Orisakwe, O.E*., Afonne, O.J., Chilaka, K.C., Obi,E., Nnamdi Azikiwe University, Nnewi Campus, Anambra State. Chromium vi and zinc ii were investigated for possible protective and/or destructive effects on murine testis. K2Cr2O7 and ZnCl2 were administered at the doses of 150ppm and 800ppm respectively in the drinking water, for a treatment period of 12 weeks. After the exposure, the testis was processed for histological examination, and epididymal sperm number counted. Zinc ii had no significant difference in the body weights and epididymal sperm numbers compared to the control. Chromium vi decreased significantly ($p<0.05$) the body weights, and epididymal sperm number, which were slightly increased when the two metals were co-administered, but yet significantly different from the control. The histologic study showed that about 75% of the Zinc-treated animals retained their normal testicular structure, while about 55% showed a normal structure in the Zinc/Chromium-treated group. Testis from the chromium-exposed group exhibited hyperplastic tubles, focal disintegration, and occasional calcification of inspissated spermatozoa in some tubules. Disintegration of spermatocytes, leading to spermatogenic arrest were evident in some tubules. Hexavalent chromium has a destructive effect on mouse testis, and this effect can be protected by Zinc ii.

PTA162 *In vitro* Bioassays in the Assessment of Ah-Receptor and Estrogen Receptor Mediated Activity in Water, Suspended Solids, and Sediments from Korean Coastal Areas. Lee, K.T.*, Seoul National University, Korea; Villeneuve, D., Kannan, K., Giesy, J., Michigan State University, Michigan, USA; Koh, C.H., Seoul National University, Seoul, Korea. *In vitro* bioassays using recombinant H4IIE rat hepatoma and MCF-7 human breast cancer cells were performed for the evaluation of TCDD-like toxicity and estrogenicity of sediments from Korean Coastal Areas. Recombinant cells have firefly luciferase gene under control of dioxin- or estrogen-responsive DNA enhancer element. Stimulation of expression of this gene by compounds acting through Ah R- or Estrogen receptor-mediated mechanism could be detected by luminescence. Twelve water samples, 5 suspended solids, and 93 sediment samples were collected, extracted with methylene chloride, S-removed and analyzed. Bioassay-derived toxic equivalents as TCDD-EQs and Estradiol equivalents (E_2-EQs) were compared with those of the values derived from instrumental (GC-MS and HPLC) analysis for TCDD-like as well as estrogenic compounds. High activities were measured for sediment cores and surface sediments. Instrumental analyses revealed the presence of PAHs, PCBs, and organochlorine pesticides.

PTA163 *In vitro* and *In vivo* Identification of Estrogenic Compounds using a Recombinant Yeast Screening Assay and Vitellogenin Measurements in Puget Sound Flatfish. French B.L.*, Lomax D.P., Roubal W.T. and Johnson L.L., Northwest Fisheries Science Center, NMFS/ NOAA, Seattle, WA. The endocrine modulating capabilities of environmental chemicals are of increasing concern for human and wildlife populations. The presence of these compounds in marine environments raises issues regarding their potential effects on fish. To address this, we have developed techniques and strategies for detection and identification of environmental compounds that interfere with estrogen receptor signal transduction. Our approach utilized both *in vitro* and *in vivo* assays to examine the estrogenic activity of environmental compounds such as alkylphenols, phthalates and bisphenol A. The *in vitro* assay used yeast cells that express the human estrogen receptor gene and that also contain reporter plasmids carrying estrogen response elements preceding the lac-Z gene. *In vivo* estrogenic activity was assessed by measuring vitellogenin (Vtg) production in juvenile English sole (*Pleuronectes vetulus*) and rock sole (*Lepidopsetta bilineata*) injected intraperitoneally with the test compounds. Fish received duplicate doses (0.25, 0.50, 0.75 and 1.0 mM / kg) on days 1 and 4 and blood plasma was collected on day 8 and analyzed for Vtg with an ELISA using a polyclonal antibody produced against purified English sole Vtg. Both assays detected estrogenic activity in response to alkylphenols and bisphenol A, while phthalates elicited little or no detectable response in either system. The yeast assay offers rapid and inexpensive screening of environmental chemicals whose estrogenic activities are mediated through the estrogen receptor, while the Vtg ELISA shows a physiologically relevant estrogenic response. These results demonstrate the complimentary use of *in vitro* and *in vivo* assays to comprehensively identify environmental estrogens.

PTA164 Antiestrogenic Activity of Polychlorinated Biphenyl Congeners in Human Breast Cancer Cells. Gupta, M.S*; Ramamoorthy, K; Safe, S., Texas A&M University, College Station, TX. 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) and structurally related halogenated hydrocarbons bind the aryl hydrocarbon receptor (AhR) and induce multiple tissue-specific responses. Several studies have demonstrated that AhR agonists inhibit multiple 17β-estradiol (E2)-induced responses in the rodent mammary, uterus and human breast cancer cells. Nesaretnam and coworkers recently reported the estrogenic activity of 3,3'4,4'4 -tetrachlorobiphenyl (tetraCB), an AhR agonist, in human breast cancer cells and in the immature mouse. However, studies in this laboratory show that neither tetraCB nor 3,3',4,4',5-pentachlorobiphenyl (pentaCB) were estrogenic in the mouse uterine model and in contrast to the previous report these AhR agonists exhibited antiestrogenic activity. The estrogenic and antiestrogenic activity of tetraCB and pentaCB were further investigated in T47D human breast cancer cells. Cell proliferation assays showed neither tetraCB nor pentaCB (1-1000 nM) alone induced cell proliferation whereas 1 nM 17β-estradiol (E2) caused a 3-fold increase in cell growth. In contrast, the congeners inhibited E2-induced cell proliferation. Transient transfection assays were performed using the following E2-reponsive plasmids, which contained the bacterial chloramphenicol acetyl transferase (CAT) reporter gene: pCKB-CAT which contained the rat creatine kinase-B gene promoter and pCATD-CAT which contained the human cathepsin D gene promoter. T47D cells transiently transfected with PCD showed that neither tetraCB nor pentaCB alone induced CAT activity in contrast to an 8-fold induction by 1 nM E2. In T47D cells transiently transfected with pCKB, 1 nM E2 caused a 4-fold induction in CAT activity whereas tetraCB and pentaCB alone had no effect on CAT activity. However, both tetraCB (1000 nM) and pentaCB (1000 nM) significantly inhibited E2-induced CAT activity in cells transfected with pCKB. Competitive estrogen receptor ER) binding assays indicated that tetraCB and pentaCB did not bind the estrogen receptor. These data suggest that neither tetraCB nor pentaCB is estrogenic in T47D cells and these AhR agonists exhibit antiestrogenic activity.

PTA165 Development of a Model System to Test Chemical Effects on Sexual Differentiation. Papoulias, D.M.*, Environmental and Contaminants Research Center, Columbia, Missouri; Noltie, D.B., University of Missouri, Columbia, Missouri; Tillitt, D.E., Environmental and Contaminants Research Center, Columbia, Missouri. The recent focus on endocrine disrupting chemicals has led to extensive efforts to develop screening assays that detect exposure or demonstrate effects of this group of chemicals on wildlife species. However, to date, no test has been accepted that screens chemicals or environmental mixtures for effects on the primary germ cells with subsequent alteration of sexual differentiation of reproductive organs and gametes. To address this need, we are developing a model system which uses as the test organism the d-rR strain of medaka *Oryzias latipes*, a gonochoristic egg-laying cyprinodont fish. In this, the first phase of model characterization, we evaluate the effects of a synthetic estrogen. Ethinyl estradiol (EE) was injected into the embryos within 24 hrs after feritilization and before completion of epiboly. Effects on survival, fitness, primary germ cells, gonadal ducts, gonads, and secondary sexual characteristics are evaluated. Gonad histopathology indicates that a single injection of 0.5 to 2.5 ng EE per embryo can cause complete sex-reversal of genetic males to phenotypic females. Sex-reversed males had female-typical duct and secondary sex character development. No intersexes were observed.

PTA166 A Short-Term Dosing Model for Detecting the Effects of Environmental Contaminants on Thyroid Hormones in the Rat: A Preliminary Report. Crofton,K.M.*, Craft,E.S., Ross,D.J.. DeVito,M.J., US EPA, Research Triangle Park, NC. Previous reports have suggested that developmental exposure to polychlorinated biphenyls (PCBs), dioxin, and other endocrine disrupting chemicals decrease levels of the circulating thyroid hormones, triiodothyronine (T3) and thyroxine (T4). The purpose of this project is to develop and validate an animal model to test for additive or non-additive interactions of mixtures on thyroid hormone homeostasis. Initial efforts have focused on developing a short-term dosing protocol to characterize contaminant effects on serum thyroid hormones and CYP1A1 (EROD activity) as biomarkers for disruption of thyroid hormone homeostasis and Ah receptor activation, respectively. In this study, Long Evans rats (female, 27 days of age) were dosed with varying concentrations of endocrine disrupting chemicals for four consecutive days. Dose response curves were generated for the following chemicals: PCB 77, PCB 118, PCB 126, PCB 153, PCB 162, TCDD and 2,3,4,7,8-PeCDF. One day post-dosing, the animals were sacrificed and serum and livers were harvested. T3 and T4 were measured via radio-immunoassays, and CYP1A1 activity was determined from liver microsomes. All PCB congeners, dioxin, and PeCDF were found to produce abundant decreases in T4 and only slight sporadic decreases in T3. All the PHAHs, with the exception of PCB153, also caused increased EROD activity. Data collected to date suggest that this short-term dosing protocol is adequate for detecting PHAH-induced hypothyroxenemia and CYP1A1 induction. Future studies will involve dosing animals with combinations of these chemicals to determine the possibility of additive and/or non-additive effects on thyroid hormones.

PTA167 Bioassays to Validate Diets for their Use in Aquatic Toxicological Research. Kemler, K*., Duda, C.A., Blankenship, A.L., Villalobos, S.A., Kannan, K., Giesy, J.P., Michigan State University, East Lansing, MI; Hinton, D.E., University of California, Davis, CA. Despite good availability of fish diets for toxicological research, few can be described as purified, standardized and consistent when compared to alternative undefined (closed formulation) diets. Fish can be induced to certain adventitious responses by diet. Therefore, it is important to examine food preparations for the presence of contaminants and/or contaminant-related effects. A purified casein-based diet (PC-diet) has proven nutritionally adequate for the raising of medaka (*Oryzias latipes*), based on parameters such as growth and development to maturity, hatchability of embryos, viability of offspring, and normal histology (Lab. An. Sci., 42: 180-9, 1992). Our objectives were to analyze PC-diet for possible contaminants and/or biological effects *in vitro*. For chemistry, PC-diet was soxhlet extracted and analyzed (gas chromatography - mass spectrometry) for the presence of organochlorine (OC) contaminants. Additionally, its extract was bioassayed for the presence of either Ah receptor (AhR) or estrogen receptor (ER) active compounds using recombinant H4IIE-luc and MCF-7 cell lines, respectively. Chemical analyses did not show the presence of OC pesticides nor PCBs (limit of quantification < 10 ng/g), and bioassay results showed no activity in either cell line. The results indicate that PC-diet did not contain any AhR or ER active contaminants, thus validating its use as a consistent alternative to undefined conventional diets. Studies are underway to validate other diets. While PC-based diet has not been tested in fishes other than medaka, it is possible that the diet works on several other warm water species.

PTA168 Determination of Heptachlor in Corn Oil. Alice Clark, Kellie McQueen and Robert Smith. MRI, 425 Volker Blvd., Kansas City, MO. 64110. Diane Overstreet, NIEHS, 111 Alexander Drive, Research Triangle Park, NC. A method based on gas chromatography with electron capture detection was developed for determining heptachlor in corn oil at a concentration of ~ 2.74 to 14.5 µg/mL. The method, as calculated from the matrix standard curve, proved linear (correlation coefficient > 0.999), accurate (% RE of -1.4 to + 1.5%), and precise (≤ 1.0% RSD). Three dose formulation solutions in corn oil were prepared at a concentration of ~ 5 µg/mL and stored in the dark at ambient (~ 25°C) and refrigerated (~ 5°C) temperatures for 35 days. The results of the study indicated that the solutions were stable for 35 days when stored in the dark at both ambient and refrigerated temperature conditions. The results of a 3-h simulated dose study indicated no significant loss (94.7 ± 4.9 % found) of heptachlor from ~ 5 Fg/mL solutions in corn oil when stored for 3 hr exposed to ambient air and light.

PTA169 Determination of Heptachlor and Its Epoxide Isomers in Rat Blood, Milk and Tissues. Thane Westermeyer, William Sherman, Robert Moore and Robert Smith. MRI, 425 Volker Blvd., Kansas City, MO. 64110. Diane Overstreet, NIEHS, 111 Alexander Drive, Research Triangle Park, NC. Methods were developed for extracting heptachlor and its epoxide isomers (a and b) from rat blood, milk, liver, and whole pups and determining their concentrations by GC with electron capture detection. A mixture of ethyl acetate, acetonitrile and ethanol (65:15:10, v/v) was used for the extraction in blood, milk and whole pups. A mixture of hexane and ethyl acetate (4:1, v/v) was used to extract liver. Heptachlor epoxide b was well separated from epoxide a and heptachlor itself using a GC equipped with a Restek RTX-5 column and an electron capture detector. Quantitation was done using an internal standard (aldrin or isodrin, depending on the sample matrix). Standards (from 10 - 500 ng/g) were spiked into blood, milk, liver and pups, and least squares analysis performed. Correlation coefficients were 0.999, or above, for all matrices. Recoveries of standards from the blood, milk, liver and pups ranged from 90 - 100%. Heptachlor epoxides a and b were also distinguished by GC/MS. The a isomer produced more fragments than the b isomer, whose mass spectrum is dominated by peaks with m/e = 353 and 81.

PTA170 Analysis of NTP 2000 Rodent Feed and USP Corn Oil for PCB 126, PCB 153, TCDD, PeCDD and PeCDF. Robert E. Smith, Kathy Boggess, Janet Paper and John Wagner. MRI, 425 Volker Blvd., Kansas City, MO. 64110. Diane Overstreet, NIEHS, 111 Alexander Drive, Research Triangle Park, NC. NTP 2000 rodent feed and USP corn oil were analyzed for PCB 126, PCB 153, 2,3,7,8-TCDD, 1,2,3,7,8-PeCDD and 2,3,4,7,8-PeCDF. NTP 2000 feed was extracted with toluene in an accelerated solvent extractor and analyzed by high resolution gas chromatography interfaced to high resolution mass spectrometry (HRGC/HRMS). Interestingly, 2.9 ng/g PCB 126 was found. None of the other analytes were detected, though. To verify that these analytes could be detected, if present, aliquots of feed were spiked at 100 pg/g with 2,3,7,8-TCDD, PCB 126, 1,2,3,7,8-PeCDD and 2,3,4,7,8-PeCDF, and 100 ng/g PCB 153. The average recoveries of were 98.3% for 2,3,7,8-TCDD (2.54% RSD), 120.8% for PCB 126 (1.42% RSD), 53.0% for 1,2,3,7,8-PeCDD (4.89 %RSD), and 84.8% for 2,3,4,7,8-PeCDF (1.58% RSD). The recovery of 100 ng/g PCB 153 was 93.5 ng/g, or 93.5%. Corn oil blanks and spiked samples were cleaned up with silica. There was no detectable 2,3,7,8-TCDD, PCB 126, PCB 153, 1,2,3,7,8-PeCDD or 2,3,4,7,8-PeCDF in the corn oil blanks. Recoveries from spiked samples ranged from 56-120%.

PTA171 Determination of Di(2-ethylhexyl)phthalate in NIH-07 Rodent Feed. Robert Smith and William Sherman. MRI, 425 Volker Blvd., Kansas City, MO. 64110. Diane Overstreet, NIEHS, 111 Alexander Drive, Research Triangle Park, NC. A method based on solvent (acetonitrile) extraction, followed by HPLC analysis was developed for determining the concentration of di(2-ethylhexyl)phthalate in NIH-07 rodent feed over a range of ~1.38 to 4240 µg/g. The method proved to be linear (correlation coefficient > 0.999), accurate (% RE of ~ -4.1 to 5.2%) and precise (≤ 1.7% RSD). The average recovery for the matrix standard curve was 93.1%. This method was used to analyze blank feed, and 1.42 µg/g was found. An additional ~2.6 µg/g of di(2-ethylhexyl)phthalate was added to the feed, and a 35-day stability study was performed. Samples were stored in the dark at ambient (~ 25°C), refrigerated (~ 5°C), and frozen (~ -20°C) temperatures and simulated dosing conditions and analyzed using the validated method on days 0, 1, 4, 8, 28, and 35 of the stability study. The results indicated a slight trend towards loss, but feed formulation samples stored in frozen conditions were at 93.6% of their Day 0 values on Day 35.

PTA172 The Effects of Broad-band Electromagnetic Field Exposure on Mice *(Mus Musculus)*. Fernandez, V.A. *, Gagnon, Z.E., Marist College, Poughkeepsie, NY; Conetta, J.A., New York Medical College, Valhalla, NY. Twenty mice *(Mus musculus)*, strain Swiss Webster, were exposed to an electromagnetic field (12.8 V/m) continuously for a period of eight weeks. Blood smear for differentials (lymphocytes, neutrophils, eosinophils, monocytes, stabs, and nucleated red blood cells), sera for clinical chemistries (creatine kinase, alkaline phosphatase, and cholesterol), and tissue samples (the thymus and adrenal gland) were analyzed for signs of stress from the EMF. Hematology analysis showed that the immune response is gender specific. The females suppressed the neutrophils while the males suppressed the lymphocytes when exposed to the EMF. Histology analysis showed that there were gender differences with regards to the lesions in the thymus and adrenal gland. The females were adrenal sensitive while the males were thymus sensitive. Results of this experiment demonstrate the detrimental effects of electromagnetic radiation on mice and suggest possible extrapolation to human health risks.

PTA173 Long term effects of incubating in oiled gravel reduce average fitness in exposed pink salmon (*Oncorhynchus gorbuscha*) populations. Heintz, R. A.*, Wertheimer, A. C., Rice, S. D. U.S. NMFS, Auke Bay Laboratory, 11305 Glacier Hwy., Juneau, AK 99801. Long term effects on average fitness may be an overlooked consequence for pink salmon populations exposed to crude oil during embryonic development. We examined this hypothesis by incubating pink salmon in gravel contaminated with crude oil and measuring mortality: (1) during embryonic development; and (2) from emergent fry (juvenile) to adult return. Integrating the mortality estimates provided a model describing the effect of embryonic exposure on the average fitness in the exposed population. Evaluation of this model for 2 separate broods indicated a pink salmon population exposed to 15 ppb of polynuclear aromatic hydrocarbons in water will produce 40% fewer reproducing adults than an unexposed population. This reduction resulted from significantly elevated mortality throughout the life history of the fish. We also observed delayed sublethal effects, such as reduced growth of juvenile fish, and impaired homing fidelity and smaller size of mature adults exposed during embryonic development. Reduced size of females at maturity suggests further reductions in average fitness because fecundity of pink salmon is directly related to size. The results reported here are from a simulation of the exposure mechanism posited for pink salmon in Prince William Sound after the *Exxon Valdez* oil spill. We incubated embryos up to 191 days downstream from weathered oil sources whose concentrations decreased logarithmically throughout the exposure period. The effective concentration in our simulation is 100 times lower than that reported in studies based on short term bioassays involving acute exposures to unweathered oil. Because long-term effects are not detected by short-term bioassays, we conclude that short-term bioassays underestimate the risk imposed on these organisms.

PTA174 Use of Machine Vision Processing in *Ceriodaphnia* Toxicity Tests. Baummer III, J.C.*, Burlington Research Inc., Burlington NC; Ciarcia, C.A., Tardis Systems, Inc., Los Alamos, NM; Diehl, R.A., Moore, S.B., Burlington Research Inc., Burlington NC. Counting of neonates and segregation of broods in cladoceran chronic toxicity testing is by nature labor intensive and potentially subject to error depending on the biologist's expertise. To address these issues, an alternative approach to counting *Ceriodaphnia dubia* neonates in single and multiple concentration chronic toxicity tests has been developed. The system integrates a high resolution camera with good depth-of-field and a PC-based machine vision processing program written in C++. The program algorithms "train" the camera to recognize a region of interest within a cylindrical test vessel and then signal the camera to capture five consecutive images at a rate of 0.33 milliseconds per image. Initially, organisms are located in individual images; blob analysis is applied to a final composite image to establish a count based on the movement patterns. The average size of individual organisms in millimeters is calculated on the basis of screen pixels for each of the five images. A DOS data file is created with count, size and movement pattern information for statistical analysis. Evaluation of the prototype system indicates a 33% time savings per test and a 98% accuracy rate compared to visual based counts.

PTA175 Comparison Of Ecotoxicity Surrogate Tests For Assessing Wastewater Effluents. Richard A. Ferraina, Jon F. Ericson, Matthew L. Galarneau and John D. Cooney. A comparison of several toxicity test kits has been performed to identify potential surrogate screens that can accurately assess effluent toxicity. Surrogate toxicity screens should be available as self-contained kits. Kits were evaluated as to their ease of performance, robustness, end point analysis and correlation to NPDES permit species toxicity. Seven test kits were evaluated, using species from several different trophic levels. The following study gives the results of this comparison study, including advantages and disadvantages of each test, and recommendations on which test kits are most appropriate for characterizing pharmaceutical effluents.

PTA176 Antibiotic Control of Pathogen Interference in Fathead Minnow Short-term Chronic Tests. Guinn, R.J., Eastman Chemical Company, Kingsport, TN. Pathogenic interferences to fathead minnow (*Pimephales promelas*) short-term chronic tests are often observed when using receiving stream water as the test control and dilution water. The interference is recognized by large among replicate variability, non-monotonic dose response, and other symptoms. In many cases, control survival will fail to meet the test acceptability criteria, and the statistical analyses for survival or growth may falsely implicate chemical toxicity. For dischargers of non-contact cooling water that originates from the receiving stream, the use of reconstituted water as the control and dilution water may exert an even greater impact on the statistical analyses, since the hypothesis test compares the responses in the "clean" control to the test concentrations that are impacted by the pathogen. Therefore, some method of controlling the pathogenic test interference must be used to produce a valid analysis of chemical toxicity. Several methods for controlling the pathogen, such as U.V. treatment, filtration, pasteurization, etc., have been suggested by various researchers. Tests conducted with receiving stream water at Tennessee Eastman Division have historically exhibited this pathogenic test interference. During the winter of 1997/98 the problem became so severe that all tests attempted failed to meet acceptability criteria for control survival. Filtration using a 0.2 µm filter was attempted and found to be effective in controlling the pathogen, but was not utilized in tests due to the difficulty in obtaining sufficient filtrate, and the perceived possibility of toxicity removal. The addition of an antibiotic (nifurpirinol) was determined to also control the pathogen after establishing the effective dosage. A description of the pathogen interference and the results of side by side tests, with and without the addition of various dosages of the antibiotic, will be presented.

PTA177 Characterization of a Scale Disorientation Abnormality in the Pinfish, *Lagodon rhomboides*. Nye, L. B.*, RSMAS/MBF, University of Miami; Corales, J., RSMAS/MBF, University of Miami; Gassman, N. J., Broward County, Florida, Department of Natural Resource Protection; Baribeau, S., RSMAS/MBF, University of Miami; Schmale, M. C., RSMAS/MBF, University of Miami. Scale disorientation was the most common abnormality in a trawl-based survey of Biscayne Bay, a tropical estuary adjacent to Miami, Florida. Scale disorientations are patches of scales which are misaligned, recognized by changes in reflectance of the surface of the fish or by touch. Scale disorientation has been observed in several species in Biscayne Bay, most notably the pinfish, *Lagodon rhomboides* and has been reported to be more prevalent in certain areas of the Bay. 140 pinfish with scale disorientations were collected from five sites for a characterization of the abnormality. Scale disorientations were almost exclusively observed above the lateral line and occupied, on average, 9.3% of the body surface of the fish. No relationship was found between the size of the disorientation and the size of the fish. However, the size of the disorientation above the lateral line was related to the site of collection. Within an area of scale disorientation, an individual scale could be growing in any direction. "Subpatches" of scales were observed where scales nearby each other were growing in the same disoriented direction. 260 scales from normal and disorientated areas of 26 fish were evaluated for morphology. Scales from disoriented areas were more elliptical, more irregular in shape and had fewer radii. Most scales in both normal and disoriented areas were ontogenetic, based on focal distance. However, a greater number of scales from disoriented areas had a large focal distance, indicating they were replacement scales. This suggests that development of scale disorientation does not require scale loss. The cause of scale disorientations is unknown, but the greater prevalence and intensity at certain sites in Biscayne Bay suggest an environmental etiology.

PTA178 Effects of Short-Term Food Deprivation on Reproduction, Growth, and Longevity of *Daphnia magna*. Putt, A.E.*, Giddings, J.M., and Biever, R.C., Springborn Laboratories, Wareham, MA; Reinert, K.H., Keller, L.H., Rohm & Haas Company, Spring House, PA. Many herbicides and fungicides inhibit algal growth, and animals feeding on the algae may experience a temporary reduction in food supply. The link between algal production and zooplankton is often considered in ecological risk assessments of algal growth inhibitors, but little is known about the effects of short-term food deprivation on zooplankton. We subjected *Daphnia magna* of different ages to 48-hour periods of algal food deprivation and measured their reproduction, growth, and survival over 21 days. Daphnids (<24 hours old at the start of the experiment) were maintained in individual vessels, fed a measured amount of algae (*Ankistrodesmus falcatus*) daily, and transferred to clean vessels every 48 hours. Offspring were removed and counted daily. The daphnids were divided into 5 treatment groups of 10 individuals. Food was withheld from different groups on Days 0-2, 4-6, 8-10, and 12-14. The fifth group (controls) was fed daily. 48-hour food deprivation resulted in smaller broods or delayed brood release for one or two broods after the deprivation period (regardless of age at the time of deprivation), but subsequent broods were normal. Our presentation includes results of a second experiment which was extended until all of the original daphnids had died, and included measurement of the reproductive capacity of the offspring of the food-deprived animals. The results could be extended with a population model to evaluate the potential indirect effect of herbicides and fungicides on zooplankton as a function of the magnitude and duration of algistatic effect, the density and growth rate of the algae, and the density and age structure of the zooplankton.

PTA179 A Semi-Automated System to Record Time-to-Death: Constructing Hazard Models for Fingerling Trout (*Oncorhynchus mykiss*) Exposed to Cadmium. Brewer, S. K.*, DeLonay, A. J., and Little, E. E. USGS-BRD, Environmental and Contaminants Research Center, Columbia MO. Environmental toxicologists have a wealth of toxicity testing protocols from which to choose in examining the effects of chemicals on aquatic organisms and to assees the potential environmental hazard. Most studies rely on conventional techniques (e.g. LC_{50}), which are directed to meet regulatory goals and not to explain underlying principles. Conventional tests require data on the percentage of individuals that survive some predetermined duration of exposure. In contrast, survival models require data on the amount of time each individual survives a particular exposure, the time-to-death. Collecting time-to-death data is more labor intensive and more expensive than recording survival to a fixed endpoint, but statistical benefits and a better understanding of a toxicant's impact in the environment can result. We present a semi-automated video-based system that greatly increases the ease at which time-to-death data is collected. We further ran our data through a series of functions (exponential, Weibull, Normal, Log normal, Log logistic) to determine the appropriate form of the proportional hazard. Lastly, we compare our results to conventional testing as a means to assess sensitivity.

PTA180 Modifications to Environment Canada's Early-Life-Stage (ELS-)Tests Using Salmonid Fish (the "Embryo" and "Embryo/Alevin" Toxicity-test Options). Fennell, M.* and Bruno J., Environment Canada, North Vancouver, BC. Environment Canada's Biological Test Method Report EPS 1/RM/28 describes three options for early-life-stage toxicity-testing using salmonids. Laboratory studies compared the sensitivity of, and improved procedures/conditions for, both the 7-day embryo (E-)test, and the longer embryo/alevin (EA-)test, each initiated with freshly-fertilized eggs. Side-by-side ELS-test comparisons were performed along with a suite of short-term acute and chronic bioassays using negative and positive control treatments and weekly samples of BKME and CTMPE. The salmonid E-test and the 7d Fathead Minnow test were compared using potassium toxicity data. Recommendations on: species choice; fertilization and test initiation (procedure/timing); reference toxicant test performance; and EA-test duration and test temperature regime will be presented. Studies revealed that the longer-term EA-test allows for the collection of more information (i.e. not limited to the single endpoint of embryo non-viability) and more revealing evidence of adverse biological effects than the 7d E-test (e.g. abnormal development becomes obvious; similar effects occur at lower concentrations). Final recommendations for the E- and EA-test include: testing at 14 1 C, and the use of Abbott's-corrected percentages for all EC50/EC25 determinations. The E-test assesses embryo non-viability 7 days after fertilization. We recommend terminating the EA-test 7 days after ≥50 %-control hatch (test duration ~29 days) collecting observations and narrative reports of alevin-deformity and delay in hatching, and numerical records of non-viable embryos, dead alevins, and cumulative mortality for calculations of EC50/EC25.

PTA181 *In situ* Testing: Development of an Instream Chamber. Connelly, R.A.*, Goodfellow, Jr., W.L.*, Hartzell, R.S., Sohn, V.A., Bastian, M.V. and Black, J.A. EA Engineering, Science and Technology, Inc., Sparks, Maryland. An *in situ* test chamber and testing procedure were developed to be used to evaluate the chronic effects of a wastewater treatment plant effluent. Since there are no standard methods to evaluate toxicity in non-standard (*in situ*) conditions, it was necessary to develop and evaluate a procedure which could be used for future instream testing. The unique characteristics (i.e., saline water, sand bottom, shallow depth) of the effluent receiving stream were considered in the test chamber development in order to ensure a successful study design. A resident fish of the receiving stream, the channel catfish, *Ictalurus punctatus*, was selected as the test species. In order to develop a test chamber and testing protocol which met the *in situ* study objectives, four tasks were undertaken: 1. Monitor growth of test fish in order to establish test duration; 2. Design and construct a test chamber which would have adequate flow and potential exposure to stream conditions and will ensure fish survival and growth; 3. Test the chamber design and testing procedures in a simulated stream environment in the lab; 4. Conduct field validation of the chamber and procedure in a stream. A test chamber made from PVC well screen was designed with successful results. Significant fish growth was achieved in a 14 day time period. Feeding and test chamber maintenance will also be discussed.

PTA182 The Use of Photosynthetic Assays to Determine the Combined Impact of Nutrient and Toxicant Stress on Freshwater Macrophytes. Marsh, D.E.*, Chiuchiulo, A., Babcock, R., Farrar, S., and Gensemer, R.W. Boston University, Department of Biology, 5 Cummington St., Boston, MA 02215. Physiological endpoints are being used to elucidate the impacts of simultaneous nutrient limitation and toxicant stress on freshwater macrophytes. Chlorophyll fluorescence induction, a measure of the quantum efficiency of photosystem II, was used to investigate the interaction of these stressors with respect to their impact on the aquatic macrophyte *Lemna gibba*. Our goal was to use this photosynthetic assay, along with individual and population-level measurements, to determine the toxicity of different types of chemicals on freshwater plants under various nutrient-limited conditions. 10-day bioassays were conducted in media deplete of either nitrogen or phosphorus. Nutrient limited plants were exposed to either cadmium (50-250 ppm) or anthracene (0.3-3 ppm). Biomass of plants exposed to cadmium under either nitrogen or phosphorus limitation did not increase, and became severely chlorotic during the exposure. Photosynthetic measurements also indicate high toxicity even at low cadmium concentrations. Anthracene exposure produced dose-response patterns in both physical and photosynthetic measurements in both nutrient limited situations. However, plants did not become severely chlorotic as did plants exposed to cadmium. Data suggest that metals are more toxic than organics under nutrient limitation. This photosynthetic assay may be useful in determining the impact of different types of toxicants on aquatic plants under natural conditions. This would allow for a more accurate assessment of the combined ecological impact of these disparate chemical stressors on aquatic communities.

PTA183 Blue Shiner Sensitivity to PCP, Ammonia, Cadmium, KCl and SDS: How Similar Are These Endangered Fish to Fathead Minnows? Keller, A.E.,* and Millar, K., USEPA, Athens, GA; Maudsley, J. and Dorn, L., Mantech Environmental, Athens, GA. The state of Alabama is home to a number of endangered or threatened aquatic species. One such species is the blue shiner (*Notropis caeruleus*) that once inhabited the Cahaba River but has since been extirpated. Local, state and federal agencies are exerting a coordinated effort to study and improve the quality of blue shiner habitat in the Cahaba River. It is hoped that this information will permit the successful reintroduction of this and other fish species, such as the goldline darter and Cahaba shiner, into this once faunally rich area. Seven-day laboratory toxicity tests were performed with blue shiner and fathead minnow larvae to compare the applicability of fathead minnow-related water quality criteria to the blue shiner. In side-by-side tests of sensitivity to SDS, ammonia, KCl, cadmium and PCP, there were no differences in the LC_{50}s of these two species for SDS, cadmium or PCP. Blue shiners were slightly more tolerant to ammonia than were fathead minnow larvae, but the shiners were less tolerant to KCl. Growth effects were more subtle.

PTA184 Effects of alcohol and formalin (as preservatives) on measurements of weight and projected area on *Neanthes arenaceodentata* (Polychaeta). Duke, B.M, ASci Corp., Vicksburg, MS; Moore, D.W., U.S. Army Corps of Engineers, Vicksburg, MS; Farrar, J.D., ASci Corp., Vicksburg, MS. When evaluating growth in bioassays, logistics often dictate preservation of animals for subsequent measurements. Yet, relatively little is known concerning the effects of preservation on the measurement of growth. The effects of various combinations of ethanol and formalin preservation and storage time on growth (projected area and wet weight), in the marine polychaete *Neanthes arenaceodentata*, were assessed. Three treatments were evaluated: 1) preservation in ethanol alone (ethanol), 2) ethanol for two weeks followed by formalin for two weeks (ethanol / formalin), and 3) formalin alone (formalin). Animals were measured initially (day 0), and incrementally thereafter (days: 1,5,7,14,21,28). Projected area measurements were made using image analysis; weight was measured as wet weight. Significant changes were observed in the measured size of test organisms over time and across preservation type. Measurements of area at day 28 increased (relative to day 0 measurements) by 7.2%, 4.4%, and 21%, respectively, for the ethanol, ethanol / formalin, and formalin only treatments. Weights of animal preserved in ethanol only and formalin only for 28 days decreased by 28 and 29%, respectively. However, weights of animals in the ethanol / formalin treatment actually increased (12%) relative to day 0 measurements. Projected area measurements in the formalin only treatment increased slightly over the 28 day period, while area measurements for ethanol / formalin and ethanol only treatments remained constant. Weights of animals preserved in ethanol only and formalin only decreased over the 28 day period, while weights of animals preserved in ethanol / formalin increased. Results of this work are discussed in terms of implications for the evaluation of growth in chronic sublethal bioassays.

PTA185 The Use of Microorganisms for Toxicity Monitoring the Surface Water of Baikal Region. Barkhutova D.*,Namsaraev B., Molotov V., Bryanskaya A. Institute of General and Experimental Biology of Siberian Division of the Russian Academy of Sciences, Ulan-Ude, Russia. The most widely-distributed toxic organic contaminants of surface water of Baikal Region are oil-products, phenol, surface active agents, dioxins and other xenobiotics and faecal pollution. The purpose of our study is the toxicity monitoring of the surface water of Baikal region by indicator microorganisms. The samples were taken from the upper horizon water-level in June - August, October 1997 in South Baikal, in Chivirkuisky Bay and the Selenga river. In the Selenga river the number of saprophytic microorganisms was 5,400-46,000 cell/ml, in South Baikal and in Chivirkuisky Bay - 5,000- 43,000. The highest number of saprophytes was observed in the period of summer high water in June and it correlated with the organic contaminant. It was revealed in the vicinity of Ulan-Ude, below Selenginsk paper mill, that was indicated of the anthropogenic character of the pollution. Cellulolytic aerobic bacteria were found only in Chivirkuisky Bay, where submerged plants were spreaded - 1-10 cell/ml, anaerobic bacteria - 1-10 cell/ml. Also anaerobic cellulolytics were detected in Selenga at the site of Selenginsk paper mill sewage disccharges. In the more pure water of Selenga the number of hydrocarbon-oxidizing microorganisms was 1-10 cell/ml, in polluting place of the river it was reached up to 10,000 cell/ml, in South Baikal, in Chivirkuisky Bay - 10-100 cell/ml. Coliform-index of surface water was the most high value in Selenga-River near by Ulan-Ude - up to 1380 cell/l. Cyanobacteria generating toxic agents were detected in testing simples. Microcystis aeruginosa Kuetz. Emend. Elenk, Anabaena lemmermanii P.Richt, Gleotrichia echinulata (J.E.Smith) P.Richt, Gleotrichia pisum (Ag.) Thur, Goelosphaerium kuetzingianum Naeg, Anabaena scheremetievi f.scheremetievi Elenk, Aphanizomenon flos-aquae (L.) Ralts, Anabaena flos-aquae f.flos-aquae (Lingb.) Breb. have identified.

PTA186 Utilizing a robust and responsive *In situ* bioassay to predict ecologically relevant consequences of toxic stress. Maltby, L.*; McLoughlin, N.J.; Wood, R.; Clayton, S.A., University of Sheffield, Sheffield, U.K.. The effects of complex discharges on receiving water quality can be assessed and monitored using *in situ* bioassays. The objectives of this study were to investigate the robustness (i.e. variability), responsiveness (i.e. magnitude and generality of response) and ecological relevance of a *Gammmarus pulex* feeding rate *in situ* bioassay. Sampling stations were established upstream ('reference') and downstream ('contaminated') of point discharges causing known ecological impacts. Major contaminant types associated with the selected discharges included: heavy metals, surfactants, pesticides, disinfectants, organic enrichment and chlorine. Male gammarids were deployed in individual cages for 6 d and feeding rates calculated. The macroinvertebrate community was sampled and leaf litter processing rates determined. Robustness was high across all reference stations with the bioassay demonstrating relatively low variability for an *in situ* test (C.V.< 35%). *Gammarus* feeding rate was significantly lower at contaminated stations with the magnitude of feeding rate depression ranging from 20 to > 90%. Power to detect significant changes in response at downstream stations was high even for relatively small changes in feeding rate. Community structure indices and leaf litter processing rates were positively correlated with feeding rate. *Gammarus pulex* is a key "shredder" in many lotic systems and a mechanistic model is proposed that links long-term ecological responses with short-term changes in feeding rate of this key species. In conclusion, *Gammarus* feeding rate provides a robust and general measure of water quality, and the *in situ* bioassay provide a general measure of stress that is indicative of ecologically significant effects.

PTA187 The Effects of Temperature on the Sensitivity of Two Novel Test Species, *Hydra attenuata* and *Lophopodella carteri*: *In situ* and Laboratory Studies. Lavoie, D.R.* and Burton, G.A., Jr., Wright State University, Dayton, OH. Temperature is an important abiotic stressor when assessing the toxicity of temperate fresh water environments. Cold and warm temperature extremes may increase the sensitivity of invertebrate species to chemical stressors. As part of *in situ* methodology development the hydra, *Hydra attenuata*, and the bryozoan, *Lophopodella carteri*, were used in the laboratory and in field deployments (*in situ*) to assess multiple stressors. Each test species was exposed in treatments of potassium chloride (KCl) and ammonia chloride (NH_4Cl), at temperatures ranging from 5-35°C. Results imply that at temperature extremes (*i.e.* below 10°C and higher than 30°C) sensitivity to chemical stressors changes. *Hydra* and *Lophopodella* responses were compared to other test organism responses (*e.g. Chironomus tentans*, *Hyalella azteca*, and *Pimephales promelas*). Determination of temperature tolerances is useful for understanding when to initiate seasonal *in situ* monitoring (*i.e.* early in the spring and late into the fall) and for understanding natural and anthropogenic stressor interactions.

PTA188 Toxicity tests with different age groups of *Mysidopsis juniae*. Prósperi, V.A*.; Bertoletti, E.; Buratini, S. V. CETESB - Environmental Sanitation Technology Agency, São Paulo, SP, Brazil. Several methods for toxicity test with marine mysids species have been described in the literature. Since 1989 CETESB is using a mysid shrimp, *Mysidopsis juniae*, with the purpose of conducting acute toxicity test. The juveniles obtained from our laboratory stocks were cultured in natural filtered seawater (salinity 34±2‰, pH 7,8-8,3), maintained at 25±2 °C with a photoperiod of 16:8 light:dark and fed with enriched *Artemia* sp nauplii. In order to test whether organisms with different ages could be used in tests instead of those of 3 days (standardized protocol), sensitivity to zinc sulfate was determined for different age groups: 1 to 8 days old as well as for pools with different age ratio, ranging from 1 to 4 days old. Aditional experiments with sodium dodecyl sulfate and potassium dichromate were also conducted. The results showed that the sensitivity didn't vary significantly between different age groups (factor < 1,3) and were similar in terms of analytical precision to the method with 3 days old organism (factor < 1,2). It was observed that organisms with mixed age between 1 to 8 days could be used in toxicity tests. In practical terms, the laboratory routine determined that organisms between 1 to 4 days old should be used.

PTA189 Concentrating ambient water samples for toxicity testing. deBruyn, A.M.H.* and Rasmussen, J.B., McGill University, Montreal, PQ. The toxicity of most Canadian waters is too dilute to elicit a response from even very sensitive bioassays. Several methods are available to "boost" a weak toxic signal, but extraction methods have been criticized as ecologically irrelevant and concentration methods are prone to substantial artifacts (as major constituents are concentrated along with trace contaminants). We are developing a method to concentrate trace contaminants but maintain background levels of major natural constituents of ambient waters. Samples concentrated in this way may be tested with well known and reliable bioassays. This approach provides a quantitative estimate of the level of toxicity present in a water body, in the context of the actual ambient conditions.

PTA190 Determining Acceptable Range of Toxicity Testing Sensitivity Response. Cuello, R*, Hibler, L.F., Word, J.Q., Battelle/Marine Sciences Laboratory (MSL), Sequim, WA. Using LC_{50} data and typically derived no-observed-effect-concentrations (NOEC) to assess comparability of toxicity tests does not appear to provide sufficient sensitivity. The MSL has conducted sediment toxicity tests with a variety of species for a number of years involving control survival and reference toxicity tests. Previous reference toxicity results with *Rhepoxynius abronius* indicate that NOECs calculated using a model based on first derivatives and Sigmoid curves may be a better method than typical NOEC calculations when determining the acceptable range of sensitivity response. These investigations included performing reference toxicity tests with more concentration series at the lower concentration end where percent survival decreases rapidly. The test results also indicated that the use of NOECs from traditional reference toxicity tests without numerous lower end concentrations does not produce assurance of test validation. Therefore, more low level effects based contaminated concentrations need to be run. In order for toxicity test results to be interpretable, we will be investigating areas to determine the effects of sensitivity in relation to toxicity test validation. This includes 1) performing similar studies with other species to see how other organism sensitivity compares with *Rhepoxynius abronius* sensitivity; 2) running a test with two different populations of *Rhepoxynius abronius* with multiple concentration series at the lower concentration end and compare NOEC's using the first derivative to determine population sensitivity; and 3) determining where in the slope of a Sigmoid curve an NOEC value should be derived from. The results of these tests will help determine the acceptable range of sensitivity response so that toxicity tests are more interpretable.

PTA191 The Relationship of Fluctuating Dose on Chronic Toxicity of Ammonia. Goodfellow, Jr., W.*, McCulloch, W., Sohn, V., Bastian, M., Koerber, A. and Christensen, L. EA Engineering, Science and Technology, Inc., Sparks, Maryland. Most chronic toxicity tests are performed with a constant dose experimental design. However, many effluent discharges have a fluctuating concentration of compounds in the wastestream. The chronic toxicity of ammonia was evaluated to the fathead minnow, *Pimephales promelas* using 7-day short-term chronic test exposure strategy. Experiments with constant dosage and pulse dosage experimental designs were employed to evaluate whether a period of less ammonia allowed the organisms to tolerate high doses of fluctuating ammonia concentration. Ammonia was selected as the toxicant because of evidence in the literature suggesting that acute toxicity of ammonia was less for pulse-doses and the fact that ammonia is a common constituent of most municipal effluent and its mode of toxicity is rapid. The results of the two exposure regimes were compared against each other as a freshwater discharge and as various concentrations of effluent ionic strength. The results indicated that when compared as a constant concentration to the maximum pulse dosage, the fathead minnows were able to tolerate concentration of approximately 2 times the constant dosage.

PTA192 **Determination of Glutathione Content in Marine Fish.** Hussain*, S., Khan, A. T., and Atkinson, A. College of Veterinary Medicine, Nursing and Allied Health, Tuskegee University, Tuskegee, AL. The present study was conducted in marine fish species to evaluate the relationship of glutathione (GSH) levels and their living environment. Since, GSH, a ubiquitous sulfhydryl compound is known to play an important role in protecting against various chemicals, it is interesting to know if there are any alterations in GSH levels in different species living in different habitats (bottom feeders vs. non bottom feeders). To determine the levels GSH in marine fish, Spanish mackerel, Flounder, Striped bass, sheephead, and Croaker were collected from the Pensacola Bay, Florida. Fish were frozen and brought to the laboratory and liver tissues were dissected and analyzed for GSH content as measured by the Ellman method. The highest level of GSH was found in Spanish mackerel while the lowest level was found in Striped bass. The decreasing order of GSH content was, i.e., Spanish mackerel > Sheephead > Croaker > Flounder > Striped bass. The higher levels of GSH content suggest that the Spanish mackerel may have been subjected to toxicant exposure. The lowest level of GSH content in Striped bass suggests that this fish may have been exposed to lower levels of toxic chemicals. In conclusion, the results suggest that the changes in GSH levels may be biomarker for toxicant exposure in marine fishes living in different environmental conditions.

PTA193 **Impact of Road Salts on an Urban Watershed.** Schroeder, J.E.*, Beak International Incorporated, Brampton, ON, Solomon, K.R, Canadian Network of Toxicology Centres, University of Guelph, Guelph, ON. Public pressure for the maintenance of highway conditions in winter, as well as the legal liability of the provincial government for unsafe driving conditions, have resulted in a steady increase in the application of salts to Toronto area roads. To determine whether applications of road salts are likely to impair aquatic species, a probabilistic risk assessment was carried out on the Don River, which flows for approximately 10 km of its length beside one of Toronto's busiest highways, the Don Valley Parkway. Chloride data, collected during 30 years of monitoring in the Don River, were provided by the Ontario Ministry of Environment and Energy and acute toxicity data were obtained from the AQUIRE database. Three sites were selected for study, representing probable low, high and moderate impact areas. Because increases in chloride concentrations were consistently observed during the winter and early spring months, the period from November to April was selected as the most probable worst case chloride concentration period. For each site, November to April data for chloride concentrations, collected during the 1990s, were ranked and plotted, along with toxicity to aquatic organisms, as cumulative frequency distributions. From the plots, the probability that concentrations of salts would exceed effect concentrations for a number of species were determined. In addition, a trend analysis was carried out using 30 years of winter chloride data from the Main Don River site to determine whether levels of chlorides are increasing in the Don River. The data indicated that, for the Main Don River site only, winter chloride levels could exceed the concentration expected to affect a significant proportion of species. Trend analysis did not indicate increasing concentrations in the Don River.

PTA194 **A Comparison of the Effects of Salinity and Chloride Ion on the Bioaccumulation of Cadmium in *Menidia beryllina*, the Tidewater Silverside.** L.J. Schuler* and M.G. Heagler. Department of Biological and Environmental Sciences, McNeese State University, Lake Charles, Louisiana, USA. It is well known that cadmium accumulates internally in fish during waterborne exposure. Water chemistry determines which species of cadmium are dominant in each system and speciation of metals in water determines their bioavailability. In waters with high chloride concentrations, cadmium will form complex ions with the available chloride ions. In previous work, *Menidia beryllina*, tidewater silverside, was exposed to 35 μg/L cadmium with varying salinity from 0‰ to 20‰ for a 29 day exposure period. The organisms at the lowest salinity accumulated the greatest amount of cadmium (7.73 μg/g). In contrast, the organisms at the highest salinity accumulated the least amount of cadmium (1.50 μg/g). This relationship is consistent with the MINTEQA2 Model version 3.10, where at 0‰ salinity 11.3% of the cadmium is complexed as $CdCl^+$ and at 20‰ salinity where 51% of the cadmium exists as $CdCl^+$, 32% as $CdCl_2$(aq), and 6.9% as $CdCl_3^-$. These cadmium complexes are considered unavailable for uptake in fish. The present study has been undertaken to strengthen the hypothesis that the cadmium-chloride complex is the major mechanism controlling uptake across gill membranes as predicted by the MINTEQA2 model. The organism, *Menidia beryllina*, was exposed using a flow-through proportional dilutor following the same protocols used in the previous study with the exception of varying only the chloride ion by the addition of NaCl. The dilutor system was used to deliver the seven different concentrations of NaCl from 0 - 4.5 g/L Cl^- while maintaining 35 μg/L cadmium to determine the accumulation/depuration kinetics.

PTA195 **Effects of Nutrients on Amphipod Responses to the Insecticide Heptachlor Epoxide.** Walsh, JT, Boston University; Kustron, S, Boston University; Kaufman, L, Boston University; Gensemer, R*, Boston University, Boston, MA. Our aim is to predict ecological effects of the combined stresses of nutrient enrichment and insecticide applications in aquatic systems. Previous studies have indicated that detritivores are usually nitrogen limited in natural systems, and that diet can affect an organism's ability to cope with toxic insults. We tested the responses of the amphipod *Hyalella azteca* to the toxicity of Heptachlor Epoxide (HE) under the influence of nutrient limited food supplies. *H. azteca* were raised in two groups on mixed algae from either N-limited or P-limited lab cultures. Acute toxicity experiments were used to determine LC_{50} for HE in each of the two groups. Preliminary results indicate that *H azteca* have a higher LC_{50} for HE when provided with a higher N:P ratio in their diet. These results indicate that eutrophication should be taken into account when assessing the impacts of organochlorines.

PTA195A **Temporal and spatial distribution of *Enterococcus* in sediment, shellfish tissue and water in proximity to stormwater outfalls in the Whangateau Harbour.** De Luca, S.*, Lewis, G.D., Creese, R.G., University of Auckland, Leigh Marine Laboratory, PO Box 349, Warkworth, New Zealand. The faecal bacterial group *Enterococcus* was used to trace stormwater discharges in the Whangateau Harbour (90 km north of Auckland). Three experiments were conducted; a) temporal variations in bacterial levels in shellfish tissue, b) effect of rainfall on bacterial levels in shellfish tissue, sediment and water, c) spatial distribution of bacteria around a stormwater outfall. A strong seasonal pattern is evident in *Enterococcus* levels in samples of both *Austrovenus stutchburyi* (cockle) and *Macomona liliana* (wedge shell) from Lews Bay (reference site) and Point Wells (putative stormwater impact site), with higher levels during winter months. Shellfish, sediment, stormwater and harbour water were sampled daily at the same two sites, during a period of rainfall and during a dry period, for 14 and 12 days respectively. A significant increase in *Enterococcus* in stormwater was associated with a rainfall event, and harbour water was similarly affected the day following rainfall. Cockle tissue at Point Wells was shown to contain significantly elevated levels of bacteria the day following rainfall, with sediment and wedge shell tissue affected to a lesser degree. Samples taken over the dry period indicated low bacterial levels in all media, excluding cockle tissue at Point Wells. It is likely that stormwater discharges are frequently affecting the quality of cockle tissue at Point Wells. Sediment and cockle tissue were sampled at various distances away from the stormwater outfall at Point Wells. Results indicate that levels significantly increase with proximity to the drain. These data further confirm that stormwater discharges into an enclosed harbour can have a detrimental impact on environmental quality.

PTA196 **An Approach to the Assessment of Environmental Fate and Effects with Minimal Ecotox and Properties Testing.**.Mohs-Davis, J.*, Pace Analytical Services, Inc., Minneapolis, MN. Purdy, R.E. and Reiner, E.A., 3M Company, St.Paul, MN. The 3M Environmental Laboratory has developed an environmental assessment approach for formulated products that estimates their fate and effects and qualitatively assesses risk. Wherever estimation is a reasonable alternative, this approach can reduce, and in some cases eliminate, the time and expense of laboratory testing. For estimations of biodegradability and bioaccumulative potential, this includes analysis of individual components using QSARs (Qualitative Structure Activity Relationships). For toxicological effects to aquatic organisms, an additive toxicity program based on the toxicity of individual components is utilized. This program generates results to within a predicted ±25% of the laboratory value. This program assumes that any compositional contributions to the overall toxicity of the formulated product are additive. This 3M environmental assessment process, when appropriate, also considers the fraction of a product's toxicity likely to be removed in wastewater treatment. This estimative approach to assessing environmental impact has proven to be a useful tool for addressing several business needs including customer inquiries, product development and reformulation. This poster outlines the assessment approach currently in use and under continuing development by 3M's Environmental Laboratory. The poster demonstrates the utility of this approach by examining three formulated products with different physiochemical characteristics.

PTA197 A QSAR study with aromatic hydrocarbons and a freshwater fish specie. W. Di Marzio, J. Alberdi*, M. Sáenz and M. Tortorelli, Ecotoxicology Research Program, Dept. of Basic Sciences, National University of Luján (Bs. As., Argentina). A QSAR study was performed with the following aromatic hydrocarbons: benzene, toluene, ethylbenzene, o-xylene, m-xylene, p-xylene. isopropylbenzene, n-propylbenzene and butylbenzene. Acute toxicity was evaluated on freshwater fish Phalloceros caudimaculatus, a native temperate Poeciliidae, by bioassays following the OECD (1981) guidelines with modifications. These were done in order to test volatile compounds through static assays in hermetic vessels. Total water volume was calculated to ensure a normal dissolved oxygen concentration in each vessel during 96 h. LC50's-96h were determined using the HPLC measured concentrations after 96 h of exposition. Quantitative analyses were carried out on Varian 5000 HPLC equipment under the following conditions: C18 reverse-phase column (MicroPak MCH-5, length = 15 cm); eluting mixture, methanol:water (3: 1) flow rate, 1 ml/min, volume injected: 10 (l except for butylbenzene (50 (l). Data were fitted according Hansch's approach: log 1/LC50 = log Kow k1 + k2. We obtained the following equation: Log 1/LC50 = log Kow 0.39 - 2.31. k1 and k2 were similar to those showed for others Poeciliidae as P. reticulata and C. decemmaculatus. Effects of non polar narcotic compounds, Fergunson's principle and fish physiology were discussed.

PTA198 A Nonlinear QSAR Method Based on K-Nearest Neighbor Principle and Genetic Algorithm Optimization. Ethiraj, G. S.*, Zheng, W., Cho, S. J., Tropsha, A., Laboratory of Molecular Modeling, Division of Medicinal Chemistry and Natural Products, School of Pharmacy, University of North Carolina at Chapel Hill, Chapel Hill, NC 27599. A novel automated variable selection QSAR method, based on the K-Nearest Neighbor principle (KNN-QSAR) has been developed. The KNN-QSAR method explores formally the active analog approach, which implies that similar compounds display similar profiles of pharmacological activities. The activity of each compound is predicted as the average activity of K most chemically similar compounds from the dataset. The robustness of a QSAR model is characterized by the value of cross-validated R^2 (q^2) using leave-one-out cross-validation method. The chemical structures are characterized by multiple topological descriptors such as molecular connectivity indices or atom pairs. The chemical similarity is evaluated by Euclidean distances between compounds in multidimensional descriptor space using only a subset of descriptors (topological pharmacophore). This subset is first selected randomly and than optimized via genetic algorithms. The application of the KNN-QSAR method to 58 estrogen receptor ligands as well as to several other groups of pharmacologically active compounds yielded QSAR models with q^2 values of 0.6 or higher. Due to its relative simplicity, high degree of automation, and non-linear nature this method could be potentially applicable to very large datasets of biologically active or toxic compounds.

PTA199 Fungal Biodegradation of Abietic Resin Acids Contained in an E_1- Pulp Bleaching Effluent. Soares, C. H. L. * & Durán, N. ** Dept of Biochemistry, CCB, Universidade Federal de Santa Catarina, C. P. 5079, CEP 88040-970, Florianópolis, SC, Brazil * and Instituto de Química, Biological Chemistry Laboratory, Universidade Estadual de Campinas, C. P. 6154, CEP 13083-970, Campinas,SP, Brazil. Resin acids constitute the major wood extractives contained in the pulp and paper mill effluents, which are known to be sticky materials which are difficult to remove by washing and may generate undesirable sticky deposits on process equipment. In addition, they have been pointed out as a powerful toxic constituent in many pulp mill effluents. Resin acids such as dehydroabietic, abietic, neoabietic and palustric were identified by GC-MS in the E_1-effluent, and their biodegradation by the fungus *Trametes villosa* was studied by GC-MS and reverse phase and exclusion HPLC. About 400 mg of wet weight mycelium from 7-10-day culture were incubated with 125 ml of the effluent, in 500 ml shake flasks, (the initial absorption value was 0.80 measured at pH = 7.6 at 465 nm) at initial pH 9.5 and 25 °C and mantained under agitation, at 150 rpm. The ethyl acetate extract of effluent was concentrated under reduced pressure, and the resin acids in the residue were per(trimethylsilylated), and analyzed with a Shimadzu GC-MS 2000 chromatograph. The temperature program was 3 min. at 90° C, 10°C/min. to 250° C, and 15 min at 250° C. A SE-30 fused-silica capilary column (25 m x 0.32 mm I.D.) was employed. The resin acids contained in the effluent were detected in the 0.5-17.0 mg/l-concentration range. Analysis by size exclusion HPLC revealed that these compounds occurred in a complex molecular structure, which presented molecular mass in the 900-1,100 DA range. The fungus, *Trametes villosa* degraded dehydroabietic resin acid (90 % degradation ratio in 80 h), and neoabietic (80 % degradation rate) efficiently, rather than palustric, which presented a persistent feature (35% degradation ratio), at the same period. The caracterization of biodegradation products is in progress.

PTA200 Identification of Unique Transformation Products of Organophosphorus Pesticides Under Conditions of Long-Term Storage. David, M.D.*; Li, Q.X., University of Hawaii, Department of Environmental Biochemistry, Honolulu, HI. A chemical storage building at the University of Hawaii's experimental agricultural complex near Hilo on the island of Hawaii has been designated for clean-up and removal of all chemicals which have been in storage beyond their useful life. Analysis of unlabeled storage containers for the purpose of identifying wastes for proper disposal classification has provided a unique opportunity to observe the transformation of pesticides under long-term storage conditions. Many of the storage containers were unlabeled, but those that were dated revealed that some of these compounds had been stored for 15-25 years and longer. Degradation and transformations of organochlorine, carbamate, and organophosphorus (OP) pesticides were observed in this study. OP pesticides were particularly unstable under storage conditions. In many cases, the parent OP compound had degraded entirely and identifications were made based on the presence of transformation products. Examples of OP pesticides observed with multiple transformation products include diazinon, chlorpyrifos, malathion, and dibromfos. The most common metabolites of these compounds were the expected products of hydrolysis, including the phosphoric acid and the conjugate base. Other products were also observed, including rearrangement and degradation of the hydrolysis products and the parent compound. The more highly degraded samples demonstrated 5-7 different transformation products. Samples were analyzed with mass spectrometry, utilizing GC, LC, and direct probe interfaces. The spectra of many of the observed transformation products were not included in standard spectral libraries, suggesting that they may be misidentified or remain unknown in environmental analyses. Spectra of the unique transformation products, strategies for their identification, and proposed transformation pathways will be presented.

PTA201 Comparison of Linear and BET Isotherms in Modeling of Subsurface Transport. Uchrin*, C.G., Rutgers Univ., New Brunswick, NJ; Hunter, J.G., Rutgers Univ., New Brunswick, NJ. Mathematical models of subsurface contaminant transport are often used to estimate leaching or desorption of substances from contaminated soils. Many of these soil transport models use a linear partitioning coefficient to calculate the aqueous concentration based upon solid phase concentration. The use of the linear partitioning model is predicated on assumptions that solutions are dilute, i.e., there is no limitation in dissolved or sorbed phase concentration. At high solid phase concentrations, however, this assumption is invalid, thus, erroneously high aqueous phase concentrations can be calculated (i.e. above the aqueous saturation concentration of the contaminant). Incorrect aqueous concentrations can lead to errors in the estimation of the breakthrough of the contaminant. A comparison of the behavior for a traditional advective transport model (van Genuchten 1984) is made to the same model incorporating the BET isotherm instead of a linear isotherm. Column experiments were performed using high contaminant loading to illuminate the concern. The results of both models are then compared to the column data.

PTA202 Cation-exchange Effects on Dissolved Organic Matter Collected by Reverse Osmosis from Hard, Alkaline Waters. M.B. Lovvorn* and J.S. Meyer, University of Wyoming, Laramie, WY, USA. Dissolved organic matter (DOM) concentrated through reverse osmosis membranes is used in studies of metals bioavailability and organic matter dynamics in natural waters. Although pre-treatment of supply water with sodium and proton cation exchangers typically is thought to yield similar results among various soft waters of low alkalinity, results from the two exchangers have not been compared for waters of high hardness and alkalinity. We compared the effects of different pre-treatment cation exchangers (sodium and proton) on the chemistry of DOM concentrates from hard (80 to 118 ppm as calcium carbonate), alkaline (57 to 100 ppm as calcium carbonate) waters. Without cation pretreatment, the DOM concentrate increased 5-fold in alkalinity and 4-fold in hardness, whereas dissolved organic carbon (DOC) rapidly reached an asymptote after an initial 10-fold increase. For each cation exchanger, hardness and alkalinity varied independently. With the sodium exchanger, DOC was concentrated 27-fold, hardness and pH remained about the same as background, and alkalinity increased about 10-fold. With the proton exchanger, DOC concentration increased 30-fold, pH decreased~4 units, alkalinity decreased to zero, and hardness increased 3-fold. With regard to heavy metals, major cations, and major anions, with the sodium exchanger, only sodium increased more (282-fold) than either chloride (28-fold) or DOC; with the proton exchanger, copper increased by 41-fold and zinc 1600-fold – much more than the 12-fold increase in chloride. All other heavy metals, major cations, and major anions concentrated as much or less than DOC. Thus, sodium and proton cation exchangers differ considerably in final chemistry of the DOM concentrate from reverse osmosis of hard waters with high alkalinity.

PTA203 Using Metal-Ligand Binding Characteristics to Predict Metal Toxicity: Quantitative Ion Character - Activity Relationships (QICARs). Newman, M.C., College of William and Mary, Virginia Institute of Marine Science, Gloucester Point, VA; McCloskey, J.T.*, U.S. Fish and Wildlife Service, Gloucester, VA; Tatara, C.P., University of Georgia, Savannah River Ecology Laboratory, Aiken, SC. Ecological risk assessment can be enhanced with predictive models for metal toxicity. Modelings of published data were done under the simplifying assumption that intermetal trends in toxicity reflect relative metal-ligand complex stabilities. This idea has been invoked successfully since 1904 but has yet to be applied widely in quantitative ecotoxicology. Intermetal trends in toxicity were successfully modeled with ion characteristics reflecting metal binding to ligands for a wide range of effects. Most models were useful for predictive purposes based on a F-ratio criterion and cross validation, but anomalous predictions did occur if speciation of the metal ion were ignored. In general, models for metals with the same valence (i.e., divalent metals) were better than those combining mono-, di-, and trivalent metals. The softness parameter (σ_p) and the absolute value of the log of the first hydrolysis constant ($|\log K_{OH}|$) were especially useful in model construction. Overall, metal-ligand binding theory was useful for developing QICARs for a wide range of effects and in a wide range of organisms.

PTA204 Pesticide Degradation Kinetics and Implications for Predictions of Leaching. J. Dyson*, Zeneca Agrochemicals. Simple first-order (SFO) kinetics are commonly used to describe pesticide degradation for different types of input. However, SFO kinetics do not always provide good fits to pesticide degradation data, particularly when the DT5/DT90 ratio is greater than the fixed ratio for SFO kinetics[1]. Hence, the aim of this study was to examine the relevance and constraints of using different types of degradation kinetics. Three types of degradation kinetics were considered: SF. kinetics; a generalization of SF. kinetics called First-Order Multi-Compartment (FOMC) kinetics; and finally, Hockey Stick (FOHS) kinetics. In FOMC kinetics, degradation rate in Soil, or any other environmental matrix. is taken as the average from a frequency distribution of rates characterizing degradation in a set of SF. compartments. FOMC kinetics encompass a whole range of DT50/DT90 ratios, so the "half-life" concept of SF. kinetics does not apply, where mass decreases by half for every half-life. F0MC kinetics were used because they have given better fits to data for pesticide degradation in soil after a single application or input, resulting in more accurate DT5O estimates[2]. FOHS kinetics is bi-phasic, comprising two different SF. models with a "breakpoint" in the rate of degradation, encompasses a large range of DT50/DT90 ratios and can result in more accurate DT5O estimates. While FOMC kinetics have been shown to estimate DT5O values more accurately, most leaching models use SF. kinetics to describe degradation. It is important, therefore, to test whether new forms of kinetics need to be incorporated into leaching models. This testing can be conducted for FOMC kinetics using any leaching model based on SF. kinetics, by running leaching models many times to account for the frequency distribution of degradation rates. A simple model designed for screening pesticide leaching was used for this testing to keep the computation down to practical levels[3]. The results showed that the accuracy of the predicted amount of leaching is primarily dependent on the accuracy of the DT50 value, but it is not, in general, significantly affected by the type of degradation kinetics. Thus, the key issue is to obtain accurate DT50 values irrespective of the kinetics.

PTA205 Chemical and Biological Rates of PAH Fluxes in Contaminated Sediments. Means, J.C., Western Michigan University, Kalamazoo, MI 49008 Chronically contaminated sediments represent a potential long-term source of mixtures of contaminants, exposing aquatic ecosystems to PAHs through desorption and bioaccumulation. Chronic toxicity assessments must address the rates of processes controlling the concentrations of these bound contaminants. The environmental fate of sediment-bound normal, alkylated and heterocyclic aromatic hydrocarbons were assessed in laboratory sediment microcosms incubated under controlled conditions for 30 to 180 days. Chemical analysis of accumulated residues of 62 individual PAH were conducted on thin, frozen sections of PAH contaminated sediment microcosms at time intervals ranging up to 180 days. The rates of sediment desorption fluxes for each PAH compound were determined by non-linear regression of the analytical data from each section of the microcosm cores as a function of depth at each time point. Degradation rates were estimated for each PAH compound by examaination of the changes in PAH concentrations in the lower portions of each microcosm core over the six month period. In the absence of fresh inputs of contaminants, many petrogenic PAHs are relative rapidly desorbed from surface sediments and many are degraded by naturally occurring consortia of sediment microorganisms. Supported by U.S. Dept. of Energy, the Minerals Management Service and the Louisiana Board of Regents.

PTA206 Fluxes of Selected Trace Elements Associated with Sediments in the Northwestern Gulf of Mexico. Means, J.C., Western Michigan University, Kalamazoo, MI 49008. The fate of 55 trace and rare earth elements in sediments and suspended particulates in the Mississippi River plume and coastal waters of the Northwestern Gulf of Mexico were investigated as part of a large-scale physical oceanographic study. Trace element speciation, transport and fluxes were investigated in a three-phase model in which samples (20L) are separated into a particulate (>400 nm), colloidal (1-400 nm) and dissolved phases (<1 nm) prior to elemental analysis by ICP/MS. Data were obtained on over six hundred discrete sediment samples collected on cruises over a three year period. Detection limits were 0.1 ng/ml for the dissolved and colloidal phases and ~1 ng/g for suspended particulates and bedded sediment samples. All elements were detected in all samples at trace levels or above. All analysis data were corrected for blanks and isotopic interferences. Estimates of sedimentation rates were obtained from sediment traps deployed in the region while advective fluxes were estimated using suspended particulate concentrations and current data measured by acoustic doppler methods. Suspended particulate concentrations were highly correlated with surficial bedded sediment concentrations. Estimates of the flux of each element are presented in a simple box model of the northwestern Gulf waters. Advective fluxes of selected trace elements associated with sediments range over several orders of magnitude. Depositional rates of new sediments were generally much lower than resuspension/deposition processes measured for the trace elements.

PTA207 Fate and Transport of Currently Used Pesticides in the Chesapeake Bay. Liu, B.*, University of Maryland, College Park, MD; McConnell, L.L.,USDA, ARS, Environmental Chemistry Lab, Beltsville, MD; Torrents, A., University of Maryland, College Park, MD. Susquehanna River is the largest tributary of the Chesapeake Bay, providing 90 percent of the fresh water flows to the upper half of the Bay and 50 percent overall. The Susquehanna River watershed (area = 71,200 km^2) contains areas of intensive agricultural activity. The lower Susquehanna watershed is comprised of 47% agricultural lands. Therefore pesticide inputs from the Susquehanna River are likely an important source to the Chesapeake Bay. The goal is to characterize the temporal trends in loading rates to the Chesapeake Bay from the Susquehanna River, to determine the distribution between the dissolved and particulate phases for currently used pesticides, and to measure hydrolysis rates under ambient conditions for chlorpyrifos. Ten-liter water samples were collected at the mouth of the river every 9 days over period of one year from 2/97 through 3/98. Water sample extracts were screened for 62 currently used and organochlorine pesticides and 3 transformation products. In the dissolved-phase, 19 currently used pesticides, 9 organochlorine pesticides and 3 transformation products have been observed. Atrazine, its transformation product, CIAT, and metolachlor were found at the highest concentrations with maximum values of 350, 113, and 240 ng/L, respectively. Other agricultural chemicals consistently observed were: cyanazine, α- and β-endosulfan, endosulfan sulfate, diazinon, chlorothalonil, alachlor, simazine, and chlorpyrifos. Organochlorine compounds such as hexachlorocyclohexanes, chlordanes and DDT isomers were also detected at low concentrations (generally < 1ng/L). Concentration values in the dissolved and particulate phase will be used with average daily river flow rates to estimate loadings to the Bay. Particle concentrations, dissolved organic carbon data, pH and temperature measurements will be used with results from laboratory degradation experiments to predict the half-life of chlorpyrifos in the Susquehanna River and in the Chesapeake Bay.

PTA208 Aromatic Hydrocarbons, Organochlorine Pesticides and PCB's in Sediments from Chetumal Bay, Mexico. Noreña-Barroso, E., Zapata-Pérez, O., Ceja-Moreno, V., Gold-Bouchot, G. CINVESTAV-IPN, Merida, Yucatan, Mexico. A mass mortality of Hardhead Catfish (*Arius felis*) was observed in Chetumal Bay in 1996 and, as an effort to diagnose this mortality, the concentrations of organic pollutants in the sediments were determined. Sediment samples from 17 stations were analyzed for PAHs, organochlorine pesticides and PCBs. Median concentrations of pesticides were highest for HCHs (1.46 ng/g), DDTs (1.43 ng/g) and chlordanes (0.87 ng/g). The median concentration of PCBs and PAHs was 2.96 ng/g and 2.34 µg/g, respectively. There is evidence that the PAHs found in the sediments are pyrogenic: the concentration of 4-5 ring compounds is higher than that of the 2-3 ring compounds, and the concentration of the alkyl derivatives is lower than that of the parent compounds. Spatial distribution of hydrocarbons in the bay is different than that of chlorinated compounds, this could be related to the circulation pattern and/or different sources of pollutants. There is an input of organic chemicals from Rio Hondo, a river that constitutes the border between Mexico and Belize and that flows through a sugar cane growing area. The city of Chetumal, located close to the mouth of the river, also plays a role in the entrance of organic compounds into the bay. The presence of organic pollutants in the bay is a cause of concern, considering that it is an ecosystem shared by two countries and its importance as breeding ground of the manatee and other endangered species.

PTA209 Spatial and Temporal Distributions of PAHs and Lipids in Chesapeake Bay Sediments. Murray, K.Y.(*), Canuel, E.A., Dickhut, R.M.; Virginia Institute of Marine Science, Gloucester Point, VA. The distribution of polycyclic aromatic hydrocarbons (PAHs) in sediments is often modeled based upon characteristics of the contaminant as a sorbate and the organic carbon content of the sediment. In many systems, these qualities are sufficient to predict the degree of contaminant sorption to a given sediment. However, it has become obvious in other studies that further characterization of a given system is needed in order to adequately account for sedimentary contaminant loadings. Specific surface area, mineralogical composition, and carbon quality are frequently overlooked as factors controlling the distribution of PAHs in marine sediments. The objectives of this study were to characterize lipid biomarker and PAH distributions in surficial sediments in southern Chesapeake Bay. Fatty acid, alcohol, sterol, and PAH concentrations in surface sediments were quantified in order to understand how organic carbon composition varies spatially and temporally and how organic carbon quality affects the distribution of PAHs. Lipid biomarker distributions suggest that sedimentary organic carbon in southern Chesapeake Bay is derived from a mixture of fresh and detrital phytoplankton, bacteria, and vascular plants. Phytoplankton production is the dominant factor controlling organic carbon quality; enrichments of POC, total Fatty Acids, total Sterols, and a number of biomarkers indicative of phytoplankton sources were found in samples collected in late spring and early summer. Combined lipid and PAH data indicate that organic carbon quality is an important factor controlling PAH distributions in surficial sediments.

PTA210 Metal Partitioning Between Sediment, Pore Water, and Surface Water in a San Francisco Bay Coastal Marsh. T. Campbell*, Woodward-Clyde International, Denver, CO; C. Loy, IT Corporation, Martinez, CA; and N. Navarro, United States Army Corps of Engineers, Sacramento, CA. A California EPA Phase I ecological risk assessment was conducted for a site on San Francisco Bay. The risk assessment evaluated the potential for toxicological impacts to a variety of estuarine organisms. A critical element of this risk assessment was to apply equilibrium partitioning models and associated dilution assumptions to measured bulk sediment contamination in order to predict exposure concentrations in the water column. The literature-based models and assumptions represented a major source of uncertainty in the estimates of receptor organism exposure to sediment contaminants. Sediment core samples were collected from locations in the intertidal coastal salt marsh and were analyzed for metals, organics, and physical characteristics. Sediment samples were also centrifuged to extract pore water for similar analysis. Collocated near-bottom and near-surface water samples also were collected for analysis. The resulting sediment and water data were analyzed to establish site-specific partitioning of chemicals among bulk sediment, pore water, and overlying water. The site-specific partitioning values and dilution factors will be used in the revised ecological risk assessment in place of conservative literature values.

PTA211 Seasonal Changes of Trace Metal Bioavailability in a Semi-enclosed Bay. Lacey, E. M.* and King, J. W., Graduate School of Oceanography, University of Rhode Island, Narragansett, RI. Sediment trace metal toxic influences are controlled by the degree of trace metal binding to the sediment. Surficial sediment and water column studies were performed in a semi-enclosed freshwater bay to examine seasonal changes in trace metal bioavailability. The surficial sediment was analyzed for trace metal concentrations (Cd, Cu, Fe, Mn, Ni, Pb, Zn), grain size, acid volatile sulfide (AVS) and simultaneously extracted metals (Cd, Cu, Ni, Pb and Zn) (SEMs). AVS, produced by sulfate reducing bacteria, is considered to be a reactive pool of sediment sulfide able to bind the SEMs on a molar basis. AVS has been observed to vary seasonally according to temperature of the overlying water column. In the bay, water temperature, dissolved oxygen, dissolved iron and dissolved manganese were measured. Seasonal thermal stratification was observed. Under stratified conditions, there was low bottom water dissolved oxygen concentrations and high sediment AVS concentrations. The concentration of AVS exceeded the ΣSEMs, therefore the SEM metals were considered to be bound by the sediment sulfide and not bioavailable. However, contemporaneously, manganese was remobilized from the sediments, resulting in significantly high, potentially toxic, dissolved manganese concentrations in the bottom water at the deepest station. After fall overturn of the water column, the sediment AVS concentrations decreased. Trace metals in the water column (manganese and iron) reprecipitated and were transported downslope to the sediments of the deep station. At the shallower stations, the concentration of ΣSEMs exceeded AVS indicating that the SEM metals were potentially bioavailable. At the deepest station, the AVS concentration continued to exceed ΣSEMs, however, there were extremely high (likely toxic) concentrations of manganese in the sediment.

PTA212 Hydroxyatrazine in Soils and Sediments. Lerch, R. N.*, USDA-ARS, Columbia, MO; Thurman, E. M., USGS, Lawrence, KS; Blanchard, P. E., University of Missouri, Columbia, MO. Knowledge of hydroxyatrazine (HA) sorption to soils, and its pattern of stream water contamination suggest that it is persistent in the environment. The primary objective of this research was to measure HA in soils and stream sediments. Soils with different atrazine use histories were collected from four sites, and sediments were collected from an agricultural watershed. Samples were exhaustively extracted with a mixed-mode extractant, and HA was quantitated using HPLC with UV detection. Atrazine, deethylatrazine (DEA), and deisopropylatrazine (DIA) were also measured in all samples. HA concentrations were greater than atrazine, DEA, and DIA in all field soils. Median soil concentrations were: HA, 77.5 µg/kg; atrazine, 9.89 µg/kg; DEA, 3.43 µg/kg; and DIA, 1.79 µg/kg. In sediments, HA was also present at higher concentrations than atrazine, DEA, and DIA. Concentrations of HA in sediments ranged from 10.6 to 96.4 µg/kg with a median concentration of 14.1 µg/kg. Correlations of HA and atrazine concentrations to soil properties indicated that HA levels in soils were controlled by sorption of atrazine. Since atrazine hydrolysis is known to be enhanced by sorption and pH extremes, soils with high organic matter and clay content and low pH will result in greater atrazine sorption and subsequent hydrolysis. In sediments, significant correlations of HA concentrations to organic matter, pH, and cation exchange capacity suggested that mixed-mode sorption (i.e., binding by cation exchange and hydrophobic interactions) controlled its levels. The presence of HA at the levels observed support existing hypotheses regarding its transport in surface runoff. These results also indicated an additional risk factor associated with atrazine usage is the potential impact of sediment-bound HA on aquatic ecosystems.

PTA213 A Model for Degradation of Pesticide Metabolites in Soils. Chapman, P.* Zeneca Agrochemicals This paper considers. for a single application of pesticide to the soil, a model that includes parent degradation. metabolite formation and metabolite degradation. For degradation of parent active ingredient and metabolite formation, three types of kinetics are considered: zero order kinetics in which the parent degradation is constant and independent of the amount of parent present; Simple First Order (SF0) kinetics. in which the degradation rate is proportional to the amount of parent present: and First-Order-Multi-Compartment (FOMC) kinetics in which degradation rate is taken as the average from a frequency distribution of rates characterizing degradation in a set of SF0 compartments. For degradation of metabolite. the same three types of kinetics are considered. making nine different parent, metabolite models in total. Whichever kinetics are considered, metabolite levels at time *t* can be represented generically as an integral sum of two variables multiplied together. The variables are the metabolite formation rate at any instantaneous time t^*, and the fraction of these instantaneous inputs remaining at time $t-t^*$. This sum is multiplied by the fraction of parent forming the metabolite if more than one metabolite is formed directly from parent. The metabolite degradation part of the model can be made more complex in two respects: by inclusion of a term representing delay in metabolite formation after disappearance of the parent: and by inclusion of a second term representing delay in degradation following formation. Some examples of fits of these models to real data are given. Fitting them is not straightforward owing to the large numbers of parameters, estimates of which can be inter-correlated. It was found that a sequential approach is most effective, in which the parent is fitted first. followed by the metabolite. Once fitted. estimates of metabolite DT50 and DT9O can easily be derived. This stud\' shows that degradation kinetics should be primarily regarded as pragmatic tools for estimating pesticide and metabolite degradation and environmental fate, rather than exact mechanistic descriptions of degradation. It was found that the introduction of FOMC kinetics can make significant improvements to these estimates.

PTA214 Atrazine and Vinclozolin Sorption on Soil and Sediments. Bouchard, D.C., U.S. EPA, Athens, GA. Recent research has indicated that the herbicide atrazine and the fungicide vinclozolin can act as endocrine disruptors. Since soil and sediment are important environmental sinks for both pesticides, the magnitude and kinetics of atrazine and vinclozolin sorption are important factors in determining their environmental fate and the potential for biota exposure. The objectives of this study were to evaluate the relationships between the equilibrium sorption constant (K_p) and the first-order desorption rate coefficient (k_2) for atrazine and vinclozolin. One surface soil and three fresh water sediments were used as sorbents. Data from miscible displacement studies using soil columns were analyzed using a two-domain, first-order mass transfer model to obtain K_p and k_2. In the two domain conceptualization and mathematical model, sorption is assumed to be instantaneous (and therefore at equilibrium) in the first domain, and kinetically controlled (modeled as a first-order reaction) in the second domain. The ^3H_2O used as a conservative tracer to characterize column hydrodynamics yielded breakthrough curves (BTCs) that were all symmetric and the data were described well by the advective-dispersive local equilibrium solute transport model, thus indicating hydrodynamic equilibrium during transport for the column systems. However, atrazine and vinclozolin BTCs exhibited the "tailing" that is characteristic of nonequilibrium sorption during solute transport. The equilibrium model provided poor fits for all of the atrazine and vinclozolin data except for low Kp treatments where BTC symmetry was high. However, when slow sorption kinetics were accounted for, the two-domain model simulations provided good descriptions of the experimental data.

PTA215 PCB Dechlorination in Dredged Sediments: Effect of Moisture Content. Cho, Y.-C.*[1], Sokol, R.C.[1,2], and Rhee, G-Y.[1,2], Wadsworth Center[1] and School of Public Health[2], SUNY Albany, Albany, NY. To determine whether PCB dechlorination continued in dredged sediments, congener specific analysis was performed on encapsulated sediments dredged from the Hudson River in 1978 and contemporary sediments from the dredged area. Dechlorination in the recent sediments was about 4.5 times greater than that in the encapsulated sediments, when measured as an average number of Cl's per biphenyl. The congener pattern in the dredged sediments suggested that dechlorination had stopped after encapsulation. A laboratory dechlorination assay indicated, however, that the dredged sediments still harbored dechlorinating microorganisms. To determine the effects of reduced water content, dechlorination was investigated at five different moisture levels using Aroclor 1248. When dechlorination in the sediment slurries was first evident after 8 weeks of incubation, with 9% reduction in the number of Cl's, the slurry was divided into five portions and the moisture content was adjusted to 95, 70, 45 and 15%, respectively. A portion of the original slurry served as the control. Dechlorination continued at all moisture levels with further incubation; however, the extent after 30 weeks was much greater in the slurry and 95% sediments compared to other three treatments. There was no difference in dechlorination pattern in any of the sediments. The size of the dechlorinating population in the slurry was more than one order of magnitude greater than that in sediments with the lowest moisture content (15%).

PTA216 Determination of Alkylphenol Polyethoxylates in Fish Tissue from Fraser River, B.C. by Electrospray LC/MS. Shang, D. Ikonomou, M.G. Macdonald, R.W. and Vonguyen, L. Institute of Ocean Sciences, Sidney, B.C. V8L 4B2, Canada. Raymond, B. Environmental Conservation Branch, Environment Canada, Vancouver, BC V6P 6H9. As part of our investigation of "non-conventional" contaminants in the Strait of Georgia, B.C., we have developed a normal phase LC/ESI-MS based analytical method for the determination of nonylphenol polyethoxylates (NPEOs) in sediment and sludge samples (SETAC 97. PO993). In this work a modified version of the analytical method was applied to measure the bioaccumulation of NPEOs in fish and other marine species. Determination of NPEOs in fish tissues imposes a special challenge because of the stringent requirements for sample clean-up to minimize matrix effects caused by lipids and proteins present in the bio-samples. This was accomplished with a multi-ste p sample clean-up procedure and with an enhanced chromatographic separation. Fist, the pre-dried fish tissue sample mixed with sodium sulfate was extracted with Accelerated Solvent Extractor (ASE) using a binary solvent of 1:1 hexane/acetone. Consequently, solid phase extraction and liquid-liquid extraction were performed to eliminate impurities from the extracts. Various clean-up steps were evaluated, including alumina placed in the extraction cell, gel permeation chromatography and selective liquid-liquid extraction. Liquid-liquid extraction with hexane/methanol was the most effective approach in removing the major part of the co-eluting interfering compounds from the matrix. In order to increase the *k*'s and provide more chromatographic retention for the analytes two normal phase LC columns were used in line, a NH_2 LC column was coupled to a CN column. This combination improved significantly the method specificity. The analytical method will be described and data demonstrating the accuracy, precision and detection limits of the method will be presented. The method was applied for the determination of NPEOs in fish tissues collected from the Fraser River, B.C. Comparisons of the NPEOs distribution found in the tissue samples will be made against those found in sediments collected in the vicinity.

PTA217 Polychlorinated biphenyls (PCBs) and trace elements in sediment cores from an andean and a coastal lake in central Chile. Barra, R.*, Cisternas M., Pacheco, P. and Parra O; EULA-Chile Center, University of Concepción, Concepción Chile; Sanchez-Hernandez, J.C., Focardi S. and Bargagli. R. Department of Environmental Biology, University of Siena, Siena, Italy. Data on historical trends of persistent organic pollutants along altitudinal and latitudinal gradients in the southern hemisphere are necessary to assess global distribution and deposition patterns. Chile is characterized by two parallel mountain systems (a coastal range and the Andes) and by winds constantly flowing from pacific ocean to inland. Two indisturbed sediment cores (20 cm deep) were collected from a coastal lake (Lleu-Lleu Lake, 20 m.a.s.l, 47 m deep) and in an andean Lake(Icalma lake, 1150 m.a.s.l, 135 m deep) situated at the same latitude (38° SL) in central Chile. In order to assess the deposition of persistent atmospheric pollutants, concentrations of Hg, Pb, Cd and Polychlorinated biphenyls (PCBs) were determined in the < 200 µm fraction of each 1-cm thick slices. Levels of Hg and Pb in sediments from Lleu-Lleu lake were two/three times higher than those in samples from the andean lake. However, metal concentration increased progressively in the later environment (Hg from 7.0 to 78.0 ng/g dry wt. and Pb from 2.0 to 13.3 µg/g). On the contrary, Cd concentrations were about 4 or 5 times higher in Icalma Lake sediments than Lleu-Lleu Lake. PCBs were detected only in the first 4 cm of both cores and results slightly higher in the coastal lake (< 0.5 to 5.8 ng/g dry wt) than in the Andean one (< 0.5 to 3.9 ng/g dry wt). From these preliminary results the two lakes seems scarcely affected by anthropogenic emissions of atmospheric pollutants. Outstanding results of ^{210}Pb dating and geochemical research in the two sedimentary environments will allow a better understanding of metals and PCBs deposition patterns.

PTA218 Comparative dynamics of organochlorine pesticides (OCPs) from highlands and lowlands nearby *Los Padres* pond basin, Argentina. Miglioranza, K. S. B.*, CIC, UNMP, Mar del Plata; Aizpún de Moreno, J. E. and Moreno, V. J., Depto. Cs. Marinas, UNMP, Mar del Plata; Osterrieth, M. L., CGCyC, UNMP, Mar del Plata, Argentina. The aim of this paper is to compare the adsorption and distribution of OCPs in highlands and lowlands from natural and horticultural soils. Physical and chemical characteristics (texture, humidity, pH, organic matter content) were determined in different horizons (0-15 cm, 15-30 cm, 45-55 cm ranges) and OCPs were analyzed by GC-ECD. Lindane, Heptaclor, Aldrin and DDT, including metabolites, were present. According to physical and chemical characteristics of both pesticides and soils, different behaviours were observed. Higher concentrations of total organochlorine pesticides were found in highlands from natural and horticultural soils. Fine sand and silt predominance in lowlands would largely determine the decrease on the retaining capacity of OCPs. Natural soils from lowlands had higher organic matter and clay content than farmlands. This fact would be the cause of the relatively high total OCPs content of the former. The pesticide distribution throughout soil profile resulted similar in highlands and lowlands.

PTA219 Polysulfide and Metal Speciation in Lake Porewaters. Wang, F.*, Alfaro de la Torre, C., and Tessier, A., INRS-Eau, Université du Québec, Québec, Canada. Polysulfides are sulfur polymers with a net dinegative charge. Formed by oxidation of H_2S and bisulfide ion (HS^-) and / or reaction of elemental sulfur with bisulfide ion, polysulfides are actively involved in the formation of pyrite and in the transformation of sedimentary organic matter. As S-containing ligands that fall into the "soft sphere" category, polysulfides also form strong complexes with class B ("soft acid") metal ions such as Cu(I), Ag(I), Hg(II), and Au(I), which may affect the solubility and speciation of these metals and hence their bioavailability and cycling. However, no analytical method was available to measure individual polysulfide species directly in water samples. We have recently developed a voltammetric method that is capable of measuring low levels of total zero-valent sulfur in small volumes of porewaters. The porewater polysulfide speciation can then be calculated from porewater pH and total concentrations of zero-valent sulfur and divalent sulfide, using appropriate equilibrium constants for the formation of the polysulfide species and the solubility of elemental sulfur. We used this approach to calculate polysulfide profiles with a 1-cm vertical resolution in several lakes in Québec, Canada. Results demonstrate that lake porewaters are saturated or oversaturated with respect to rhombic sulfur and that nanomolar to micromolar levels of polysulfides are present. Calculation of metal speciation indicates that metal complexes with polysulfides dominate the speciation of Cu and possibly other class B metals in lake porewaters.

PTA220 Remediation Approaches for Control of Selenium Leaching from Waste Rock Soils in the Idaho Phosphate Resource Area. Bond. M., Steinhoff P., and Möller, G., University of Idaho, Moscow, ID. Selenium leaching from the waste rock soils of mining operations in the Idaho phosphate resource area has been linked to the observation of chronic selenium toxicosis in nearby livestock operations and large-scale environmental contamination. Characterization of the reaction pathway suggests that the oxidation of seleno-pyrite to soluble selenite compounds mobilizes the surface weathered selenium. This work examines the treatment of these soils by amendments that include scrap iron granules, colloidal iron, potato processing waste, potato starch, cheese whey and Fe(III)-thermal polyaspartic acid (tpA), with and without inoculation with sulfate reducing bacteria, *Desulfatomaculum orientis* and *Desulfovibrio desulfuricans.* Additional studies examine chemical armoring of the soils with Fe(III) solution, Fe(III)–tpA solution and tpA solution. Results of the 14 to 28 d amendment studies show a >99% reduction in pore water selenium concentrations of treated soils to as low as 0.007 mg-Se/L from 3.0 mg-Se/L in the untreated soil pore water. Chemical armoring by ferric iron solution and Fe(III)-tpA solution demonstrate the ability to limit leaching to <0.0007 mg-Se/L. Field studies of these stabilization approaches are in progress.

PTA221 Nutrient and Metal Exchange across the Sediment-Water Interface in Small Polluted Lakes. N. V. Ignatieva, Institute for Lake Research Russian Acad. Sci., St. Petersburg, RUSSIA. Nutrient (P, N) and metal (Fe, Mn, Co, Ni, Cr, Cu, V, Zn and Pb) exchange across the sediment-water interface has been studied in aquatic system of small eutrophic lakes situated to the northern outskirts of St.Pe- tersburg, Russia. Very high nitrogen content is characteristic of the sediments. Dissolved nutrients were found in the sediments ma- inly in inorganic forms. Most part of sedimented nutrients is re- leased to the water column, nutrient fluxes are extremely high. It was found that less phosphates are released from the deep sediment layers by molecular diffusion. The diffusion from immediate sedi- ment-water interface and convective transport seem to be of grea- ter importance in P release. In contrast to P, most part of N is released from the deep sediment layers as ammonium-ions. The lake sediments are enriched in Mn and Pb as a result of anthropogenic pollution. Based on metal budget at the sediment-water interface in terms of sedimentation, burial within the sediment, and release to the overlying water, it was concluded that on average 40 to 80% of sedimented metals are buried. At a current stage, the sediments serve as an important source of secondary pollution and eutrophi- cation of the lakes.

PTA222 Sources and Sinks for PCBs in Upper East Fork Poplar Creek. Palmer, J.A.*, McCarthy, J.F., Southworth, G.R., Oak Ridge National Laboratory, Oak Ridge, TN. Passive monitoring of polychlorinated biphenyls (PCBs) with semipermeable membrane devices (SPMDs) was used to identify and quantify the contributions of point- and non-point sources to the total PCB flux in Upper East Fork Poplar Creek (UEFPC), a drainage system located within the Department of Energy's Oak Ridge Y-12 Plant. SPMDs were deployed at nine discharge points and seven ambient sites in the stream. The contributions of PCB sources were evaluated by estimating the PCB flux (mass per unit time) at different monitoring locations, calculated from the time-integrated estimates of the aqueous concentrations of PCBs, the volumetric flow rates of discharges, and volumetric flow in the receiving stream during the deployment period. Two outfalls together released almost 10X more PCBs than the combined contribution of the other seven outfalls. However, these point-source releases were not the principal sources of PCBs in UEFPC. The overall flux of PCBs in the stream quadrupled from 75 milligrams PCB/day at the uppermost station to 321 milligrams PCB/day at the downstream boundary of the Y-12 Plant, a net increase of almost 250 milligrams PCB/day. Point-source inputs into the stream accounted for only 100 milligrams PCB/day, showing that the principal sources of PCBs in UEFPC are from historic releases. Buried storage tanks for PCB-containing oil previously located near the stream implicated likely sources of PCBs and suggested that PCB releases may originate through diffusion of in-place contaminants from the sediment or by advection of shallow groundwater. The passive monitoring of PCB flux thus identified two discharges as the principal point sources, but clarified the significance and location of nonpoint sources associated with historic contamination.

PTA223 Release of Hydrocarbons from Untreated and Bioremediated Soils. Webster, M.T.*, Loehr, R.C., Dickerson, K.T., Beeler, M.A., Poppendieck, D.G., The University of Texas at Austin, Austin, TX. Increasingly, risk-based cleanup criteria are being used to manage the remediation of soil containing hydrocarbons. Such an approach recognizes the importance of chemical availability in assessing the potential risk posed by chemicals in the soil. Increasing evidence suggests that environmentally acceptable endpoints (EAEs) can be reached in soil even though substantial chemical concentrations remain in the soil. In addition, bioremediation processes can be used to achieve EAEs in soil. Knowledge of the potential extent and rate of release of chemicals from untreated and treated soil will greatly aid the determination of EAEs and the use of bioremediation to achieve EAEs in soil. The objectives of this research were to determine the effect of soil and chemical characteristics, and bioremediation on the release of hydrocarbons from soil. A sacrificial, batch method was used to quantify the extent and rate of release (ROR) of hydrocarbons from untreated and bioremediated field soils. The release was described using an empirical, two-site model, and the fraction of chemical readily released (F) and the rate constant describing the slow release of the remaining chemical (k_2) were determined. Results indicated that: 1) only a fraction of the hydrocarbons present in the soils was readily released from the soil into the aqueous phase, 2) F and k_2 were dependent on chemical characteristics, and 3) the extent and rate of release were dependent on the degree of prior weathering and the degree of bioremediation treatment. Such information is relevant to the determination of EAEs in soil and the use of bioremediation processes to achieve EAEs.

PTA223A Investigation of an Onsite Wastewater Treatment System (OWTS) in Sandy Soil: Part 1: Subsurface Soil and Groundwater Characterization Anderson, D.*, Ayres Associates, Tampa, FL; DeCarvalho, A., The Soap and Detergent Association, New York, NY; McAvoy, D., The Procter & Gamble Company, Cincinnati, OH; Nielsen, A., CONDEA Vista Company, Austin, TX, and Kravetz, L., Shell Chemical Company, Houston, TX. An onsite wastewater treatment system (OWTS) serving an individual home in Florida was the subject of an extensive fate and transport investigation of household cleaning product ingredients. The first phase of the study was a detailed site characterization, including site geology, soils, and groundwater characteristics. Soils at the site consisted of fine sands with less than 6% by weight silt and clay. These soils existed to a depth of at least 15 m below ground surface (bgs). Shallow soils at the water table zone had a bulk density of approximately 1.5 gm/cm^3 and a porosity of approximately 42 percent. Groundwater elevations varied seasonally from 0.5 to 1.8 m bgs. The groundwater flow direction was southwest at a horizontal gradient of approximately 0.0029 m/m. In relation to the OWTS infiltration system location, groundwater elevations resulted in unsaturated zones of 0 to 1.3 m below the system. An extensive groundwater monitoring network was installed with 90 discrete sampling points at depths from 1.8 to 9 m bgs. Aquifer slug tests and bromide tracer tests were then conducted to further define groundwater flow characteristics at the site. The saturated hydraulic conductivity was estimated at 2.5 m/day and the groundwater seepage velocity at 0.04 m/day based on these tests. The bromide plume was characterized over a 150 day period to define the impact area of a conservative element discharged to the OWTS.

PTA223B Investigation of an Onsite Wastewater Treatment System (OWTS) in Sandy Soil: Part 2: Chemical Transport Characterization Anderson, D.*, Ayres Associates, Tampa, FL; DeCarvalho, A., The Soap and Detergent Association, New York, NY; McAvoy, D., The Procter & Gamble Company, Cincinnati, OH; Nielsen, A., CONDEA Vista Company, Austin, TX; and Kravetz, L., Shell Chemical Company, Houston, TX. This phase of the OWTS study characterized the wastewater source and subsequently delineated the effluent plume for: chloride (Cl), total organic carbon (TOC), nitrogen (N), phosphorus (P), and methylene blue active substance (MBAS). Groundwater was sampled using a monitoring network consisting of 29 locations with 90 discrete sampling points at depths from 1.8 to 9 m below ground surface (bgs). Groundwater samples were recovered during peak seasonal fluctuations in groundwater level; at seasonal high water with no unsaturated soil below the infiltration system and; at seasonal low water with 0.40 m unsaturated soil below the infiltration system. Differences in groundwater quality data between the two conditions were observed. In general, measured parameter concentrations were greater under high water conditions. N was significantly elevated under both conditions; however, based on groundwater velocities determined from bromide tracer tests, the species of N at a given location appeared to be related to unsaturated zone thickness at the time the effected water was impacted by the OWTS infiltration system. MBAS concentrations greater than background extended approximately 6 m downgradient, with lower concentrations measured under low water conditions. The horizontal and vertical extent of the effluent plume under the conditions studied appeared limited to a relatively small area.

PTA223C Investigation of an Onsite Wastewater Treatment System (OWTS) in Sandy Soil: Part 3: Fate of Anionic and Nonionic Surfactants Nielsen, A.*, CONDEA Vista Company, Austin, TX; DeCarvalho, A., The Soap and Detergent Association, New York, NY; McAvoy, D., The Procter & Gamble Company, Cincinnati, OH; Kravetz, L., Shell Chemical Company, Houston, TX. Septic systems are used to treat approximately 25% of all domestic wastewater in the United States. This domestic wastewater includes the surfactants used in household cleaning products. The study reports the fate of surfactants linear alkylbenzene sulfonate (LAS), alcohol ether sulfate (AES) and alcohol ethoxylate (AE) in a single home septic system in Jacksonville, Florida, in which the leaching laterals reside in a fine sand unsaturated zone of less than 1 meter. During the wettest times of the year it is likely that effluent from the septic system passes directly into the ground water without exposure to the unsaturated zone. Groundwater was recovered and analyzed for LAS, AES and AE from two sampling events, during the seasonal high and low groundwater table levels. The first sample was during the wet season with an unsaturated zone of only approximately 0.01 m. The second sample was taken during the dry season when the unsaturated zone was approximately 0.4 m. It was clear from both sampling events that AES and AE were removed within 0.2 m of the leaching laterals. LAS did migrate 11.5 m horizontally and 3.7 m vertically. As it migrated, LAS was removed by biodegradation and adsorption. This study indicates that LAS, AES and AE are all readily removed in septic system leaching fields in situations with minimal unsaturated zones.

PTA223D Investigation of an Onsite Wastewater Treatment System (OWTS) in a Sandy Soil: Part 4: Adsorption and Biodegradation of LAS Doi, J.* and Marks, K., Roy F. Weston, Inc., Exton, PA; DeCarvalho, A., The Soap and Detergent Association, New York, NY; McAvoy, D., The Procter & Gamble Company, Cincinnati, OH; Nielsen, A., CONDEA Vista Company, Austin, TX; and Kravetz, L., Shell Chemical Company, Houston, TX. This work is part of an overall program for investigating the fate and transport of household cleaning product ingredients in onsite wastewater treatment system. The objective of this work was to determine sorption coefficient and biodegradation rate constants for incorporation into a predictive model for an OWTS. In both the sorption and biodegradation studies, three distinct soil samples were collected from a Jacksonville, FL septic tank field study site. Different concentrations of the test substance were added to a series of test vessels that contained upgradient groundwater and the collected soils. The adsorption test was designed to determine the partitioning of LAS between each soil type. Results indicated that the Freundlich isotherm coefficient (K) ranged from 0.55 -1.38, the adsorption coefficient (K_d) ranged from 0.43 - 4.02 and the rate of ultimate degradation (first order rate constant - K_1) ranged from 0.08 - 2.17 day^{-1} with increasing distance (0.3 - 1.2 m vertically and 0 - 6.1 m horizontally) from the leach field. All three soils showed that 60-80% of theoretical CO_2 was produced over the 30 day period of the test. These results demonstrate that both adsorption and biodegradation are key factors in the removal of LAS in a subsurface soil system.

PTA223E Investigation of an Onsite Wastewater Treatment System (OWTS) in a Sandy Soil: Part 5: Predictive Model for Cleaning Product Ingredients McAvoy, D.*, The Procter & Gamble Company, Cincinnati, OH; DeCarvalho, A., The Soap and Detergent Association, New York, NY; Nielsen, A., CONDEA Vista Company, Austin, TX; and Kravetz, L., Shell Chemical Company, Houston, TX. Septic systems are used to treat approximately 25% of all domestic wastewater in the United States. Because of their common use, predictive tools are needed to assess the exposure concentration of down-the-drain chemicals in subsurface environments following discharge from these systems. To meet this need, a mathematical model was developed for predicting the fate and transport of household cleaning product ingredients in septic systems. The ultimate goal was to develop a screening-level tool that could be used by chemical suppliers, product manufacturers, and government authorities, for predicting exposure. The model consists of three interconnected compartments (a septic tank, an unsaturated zone, and a saturated zone) with removal in each compartment governed by sorption and biodegradation. All of the equations used in the model are based on continuity of mass. To have confidence in its predictive capabilities, the model was tested with laboratory and field data. The mathematical model formulation and practical examples of how this model could be used in exposure assessment are presented.

PTA223F The role of bioturbation by the marine amphipod, Corophium volutator in laboratory multi-species studies. Silvana Ciarelli, Department of Ecology and Ecotoxicology, University of Amsterdam, The Netherlands; Belinda J. Kater, National Institute for Coastal and Marine Management, The Netherlands; Nico M. van Straalen, Department of Ecology and Ecotoxicology, University of Amsterdam, The Netherlands, Peter G. Schout, AquaSense consultance, Amsterdam; A. Hannewijk, AquaSense consultance, Amsterdam, The Netherlands. Single-species tests are extensively used in research, regulation and monitoring studies to provide information on the biological effects of complex chemical mixtures and to discriminate among sediments containing low to moderate levels of contamination. In addition to sediment-water partition coefficients and accumulation of chemicals in biota, these are useful tools in sediment risk assessments. When single-species are exposed to sediment-bound contaminants or/and sediment-water partition coefficients and accumulation levels are calculated on EqP model basis, two important questions remain. What amount of sediment-bound contaminants are biological available and which are the factors that influence bioavailability and toxicity of sediment-bound contaminants? More complex test systems, such as multi-species studies, which provide greater insights into the factors affecting environmental chemical transport and which take into account species interactions, are needed. Biological processes such as, sediment bioturbation and sediment resuspension by benthic organisms as a consequence of burrowing activity, are important factors which can affect the fate and availability of sediment-bound contaminants. Sediment bioturbation is known to play an important role in changes in the fate and in the cycling of sediment-bound contaminants to organims. Preliminary results of experiments carried out with two different species and focusing on the effects of sediment bioturbation by *Corophium volutator* on the accumulation of fluoranthene in mussels and worms are shown.

PTA224 The Relative Sensitivity of Four Benthic Invertebrates to Select Metals in Spiked Sediments. Milani, D.*, McMaster University/National Water Research Institute, Burlington,ON; Reynoldson, T.B., National Water Research Institute, Burlington,ON; Kolasa, J., McMaster University, Hamilton,ON. Environment Canada (NWRI, Burlington,ON) assesses sediment toxicity with the following benthic invertebrate sediment toxicity tests: *Hyalella azteca* 28-day survival and growth test; *Chironomus riparius* 10-day survival and growth test; *Hexagenia spp* 21-day survival and growth test; and *Tubifex tubifex* 28-day reproduction test. The relative sensitivities of these four benthic invertebrates to selected metals is currently being assessed by performing these toxicity tests in cadmium, nickel and copper spiked reference sediments. Metal concentrations are measured in the bulk sediment, pore water and overlying water. Both *C. riparius* and *H. azteca* are most sensitive to cadmium, exhibiting lesser sensitivity to nickel, followed by copper. *Hexagenia spp.* and *T. tubifex* are most sensitive to copper, followed by nickel and cadmium. Overall, *H. azteca* is the most sensitive organism to the metals examined, with LC50 's ranging from 19 to 23 ppm, and IC25's for growth ranging from 4 to15 ppm, and *T. tubifex* is the least sensitive organism to the metals examined, with LC50's ranging from 224 to 566 ppm, and IC25's for the reproductive endpoint (number of young/adult, number of cocoons/adult) ranging from 162 to 625 ppm. These preliminary results indicate that the organisms show differential responses to the metals examined which will aid in evaluating the source of toxicity in sediments and also demonstrates the need for multispecies testing in evaluating sediment toxicity.

PTA225 Toxicity of Un-ionized Ammonia to the Development of the Purple Sea Urchin, *Strongylocentrotus purpuratus*, and the Sand Dollar, *Dendraster excentricus*, in Sediment Pore Water and Elutriate Bioassays. Brown, R.W.*, TTEMI, San Francisco, CA; Bernhard, T.S., EFAWest, U.S. Navy, San Bruno, CA; Joab, B.M., TTEMI, San Francisco, CA; Leather, J.M., SPAWARSYSCEN, San Diego, CA. Laboratory bioassays were conducted on sediment pore water and elutriates prepared from sediments obtained near a number of U.S. Navy facilities in San Francisco Bay. Percentages of the normal development of the early life stages of the purple sea urchin, *Strongylocentrotus purpuratus*, and the sand dollar, *Dendraster excentricus* were determined in tests and used to evaluate sediment toxicity. Numerous samples had concentrations of ammonia in the range that has been shown to induce toxicity in a number of marine invertebrates. Correlations were found between abnormal development of the animals tested and unionized ammonia concentrations that were calculated from total ammonia measurements. There was strong evidence that unionized ammonia is a potential confounding factor in these tests. Methods to reduce the influence of un-ionized ammonia toxicity should be considered in tests of sediment pore water and elutriates when animals sensitive to unionized ammonia are used.

PTA226 Toxicokinetics of Fluoranthene in *Hyalella azteca* Under UV Light: Sediment vs. Water. Wilcoxen, S. E.,[1]* and Landrum, P. F.[2], [1]University of Michigan, Ann Arbor, MI, [2]NOAA-GLERL, Ann Arbor, MI. EPA Sediment Quality Criteria for Fluoranthene does not incorporate data on phototoxic effects and may, therefore, be underprotective. It is known that PAH toxicity is enhanced by exposure to UV light. Ten day water-only bioassays were conducted with *H. azteca* under three spectra: UV enhanced fluorescent, cool white fluorescent (which contains a small UV component), and gold (>500nm) light. For these spectra, the 10-d LC50s were 11 nmol/L, 68 nmol/L, and 411 nmol/L, respectively. The 10-d LR50s were 0.01 mmol/kg and 0.20 mmol/kg for UV and fluorescent light, respectively. Under gold light tissue concentrations up to 1.2 mmol/kg resulted in 80% survival. Based on the Equilibrium Partition model, 50% mortality would be expected at sediment fluoranthene concentration 4.95 nmol/gdw under UV light. Ten day sediment bioassays were conducted with *H. azteca* in ^{14}C-fluoranthene spiked Lake Michigan sediments under UV enhanced and fluorescent light. These 10-d LR50s were 0.04 mmol/kg and 0.09 mmol/kg, respectively. At lower sediment concentrations (320 nmol/gdw), where survival was >80% under both of these spectra, *H. azteca* accumulated less fluoranthene under UV light than under fluorescent light (0.03mmol/kg vs. 0.06 mmol/kg). Similar exposures under gold light gave >90% survival and body burdens of <0.5 mmol/kg at the highest concentration (1270 nmol/gdw sediment). Although UV exposure increased mortality in water-only and sediment bioassays, the effect is less pronounced in sediment exposures. This is presumably due to the burrowing behavior of *H. azteca* under UV that was not exhibited under less harsh spectra. However, greater body residues, which might be expected from greater sediment contact, were not observed under UV light.

PTA227 Ammonia Toxicity to *Hyalella azteca*. Lasier, P.J.* and Winger, P.V., USGS Patuxent Wildlife Research Center, Athens, GA. The amphipod, *Hyalella azteca*, is a recommended test organism for toxicity assessments of freshwater sediments and pore waters. However, these media often contain ammonia concentrations that elicit a toxic response. Reported tolerances of *H. azteca* to ammonia vary from 20 mg/L to over 70 mg/L. The effects of age, feeding during testing and different ammonium-anion compounds on ammonia toxicity were determined in 96-h static toxicity tests. Known-age animals (1, 3, 7, 14, and 21 d) and mixed-age animals (7 to 10 d) were exposed to dilutions of ammonium chloride. One block of these animals were fed at test initiation and after 48 h. In nonfed exposures, ammonia was significantly more toxic to 1- and 3-d old animals (LC_{50}s of 29 and 30 mg NH_4^+/L) compared to older animals (LC_{50}s between 64 and 72 mg NH_4^+/L). Within the fed treatment, ammonia was significantly more toxic to 1- and 3-d old animals than 14- and 21-d old animals; LC_{50}s ranged from 58 to 88 mg NH_4^+/L. Coefficients of variation for survival among replicates were greatest for 1- and 3-d old animals in nonfed treatments and lowest for mixed-age animals in both fed and nonfed treatments. Effects of different anions (Cl^-, CO_3^{-2}, SO_4^{-2} and CH_3COO^-) on ammonia toxicity was evaluated using mixed-age animals. Toxicity of $(NH_4)_2CO_3$ was significantly greater than that of the other ammonium salts due to an additive toxic effect of un-ionized ammonia produced by the greater solution pH. These tests indicate that multiple factors can influence the toxicity of ammonia to *Hyalella azteca*. Animals 3-d old or less are the most sensitive and require feeding to reduce survival variability to levels similar to the older ages.

PTA228 Toxicity Assessment Directly in the Field. Mowat, F.S., Shettlemore M.G., and Bundy, K.J. Tulane University, New Orleans, LA. Direct field tests were conducted in Louisiana wetlands to evaluate overall toxicity of organic/heavy metal contaminants and mixtures. Complex methodological challenges including operation without line power, equipment portability and ruggedness, effective phase separation and pollutant extraction, efficient data acquisition, and safety concerns have successfully been resolved using a dual instrumentation method. DeltaTox, a field portable bacterium-based biosensor, was used to qualitatively ascertain the presence of polluted areas. Microtox, a toxicity bioassay based on attenuation of light of a marine bacterium *(Vibrio fischeri)*, was then used to determine quantitative toxicity (EC_{50} values) after appropriate adaptation to field use. Saturated salt extraction was used to remove non-lattice bound pollutants from sediments via ion exchange. The extracted contaminants represent those that are labile and hence potentially more bioavailable to the wetland biota. Pb(II) appears to be the primary metal affecting toxicity in Bayou Trepagnier sediment In addition, possible synergistic and/or antagonistic interactions between metal and organic species were observed. Preliminary tests indicate that Microtox may react to natural humic substances. This affects overall toxicity either via intrinsic humic material toxicity or chelation that solubilizes other contaminants. Hormetic (stimulatory) effects resulting in observable DeltaTox light gains were sometimes observed. This may serve as a qualitative indicator of organics in these areas present at low levels. Overall, the DeltaTox/Microtox approach to field testing appears to be successful, rapid, and cost-effective for on site identification of pollution hotspots, and may lead to an increased understanding of potential hazards posed by heavy metal and organic contaminants at Louisiana sites of interest.

PTA229 Measuring the Chronic Toxicity and Bioaccumulation of Tributyltin-Contaminated Sediment Using the Estuarine Amphipod, *Leptocheirus plumulosus*. Pinza, M.R.* and Gilfoil, T.J., Battelle Marine Sciences Laboratory, Sequim, WA; Benson, T. Washington State Department of Natural Resources. Tributylin (TBT) contamination of marine and estuarine sediment is a serious management concern in the Puget Sound area. Current toxicity tests approved by the Puget Sound Dredge Disposal Agencies (PSDDA) may not be sufficiently sensitive to adequately assess the environmental risk caused by long-term, chronic exposure. Historically, the bioaccumulation of TBT has been difficult to interpret, because there is little information linking TBT tissue residues to toxicological endpoints, particularly to sublethal responses. The study objectives were 1) to measure the chronic toxicity of TBT and TBT-contaminated sediment, 2) to develop tissue-residue-based dose-response curves for bioaccumulated TBT and chronic lethal and sublethal responses, and 3) to compare the TBT-sensitivity of the chronic *Leptocheirus* test with that in a standard suite of PSDAA toxicity tests (*Eohaustorius, Neanthes*, and Echinoderm Larval tests). Two experiments used *L. plumulosus* to simultaneously measure both the chronic/sublethal toxicity and bioaccumulation of TBT in full life-cycle exposure to TBT-contaminated sediment: the TBT-Spiked Sediment experiment, and the Comparative Toxicity experiment. The former established concentration-and dose-response relationships under conditions where TBT was the only toxicant and its bioavailability was controlled by geochemical processes. The latter used sediment collected from Puget Sound in an area where TBT is known to occur; it allowed a comparison of the chronic amphipod test with tests already used in the Puget Sound area. All toxicity testing followed standard EPA or PSDDA protocols. TBT analysis of tissues, porewater, and sediment was conducted to determine bioavailability of TBT. The data collected in these experiments can be used to develop tissue-residue-based TBT dose-response curves for lethal and sublethal responses by *L. plumulosus*.

PTA230 Comparative Sediment Toxicity in the Marine Polychaetes *Polydora cornuta, Boccardia proboscidea* and *Neanthes arenaceodentata*, the Estuarine Amphipod *Leptocheirus plumulosus* and the Copepod *Schizopera knabeni*. Farrar, J.D.*, AScI Corporation, Vicksburg, MS; Bridges, T.S., USAE Waterways Experiment Station, Vicksburg, MS. Lotufo, G.R., AScI Corporation, Vicksburg, MS. Emphasis in sediment toxicity test method development has shifted from measuring acute lethality to evaluation of chronic sublethal effects following exposures to relatively low contaminant levels. Comparative toxicity data are necessary to evaluate the generality of toxicity information gained from a specific bioassay. Five bioassays were compared in this study. Survival and growth were measured following 14-day exposures with *B. proboscidea* and *P. cornuta* and 28-day exposures with *N. arenaceodentata and L. plumulosus.* All four species are currently subjects in the development of sublethal bioassays by Environment Canada (*B. proboscidea* and *P. cornuta*) and the U.S. Army Corps of Engineers (*N. arenaceodentata* and *L. plumulosus*). Additionally, survival and reproduction were evaluated following 14-day exposures with the infaunal copepod *S. knabeni.* The comparative effects of four field-collected contaminated sediments were examined. *P. cornuta* exhibited significant survival effects in all four sediments. Significant mortality was observed in two of the four sediments with *B. proboscidea*. *N. arenaceodentata* identified three sediments as toxic with significant mortality in one sediment and significant diminutions in growth in two additional sediments. S. *knabeni* exhibited no apparent survival effect with any of the field-collected sediments tested. Survival and growth effects with *L. plumulosus* were comparable to those observed with *Neanthes* and *Boccardia*. Comparisons using copper-spiked sediment were also conducted. Though variability in acute response to sediments was observed, the bioassays displayed similar sublethal responses.

PTA231 Comparative toxicity of contaminated sediment on *Leptocheirus plumulosus* (Amphipoda), *Neanthes arenaceodentata* (Polychaeta), and *Mulinia lateralis* (Bivalvia). Houston, J.G.*[1], Gray, B.R.[2], and Duke, B.M.[2], [1]U.S. Army Engineer Waterways Experiment Station, Vicksburg, MS.,[2] AScI Corp., Vicksburg, MS. The purpose of this study was to evaluate the comparative sensitivity of three proposed chronic sublethal assays to contaminated sediment. The tests consisted of a 10 day exposure for *M. lateralis* and 28 day exposures for *L. plumulosus* and *N. arenaceodentata*, using the standard testing protocol for each animal. The endpoints measured included, number of surviving adults, and adult growth (mg/dry wt.) for all animals, and reproduction (# offspring/surviving adult) for *L. plumulosus* only. Nine sediment treatments were used: sediment spiked with fluoranthrene (100, 250, and 500mg/l), sediment spiked with cadmium (0.25, 1.0, 2.0 SEM/AVS), and field collected sediment from Patrick Bayou, TX (10, 50, and 100%). Results indicated that *L. plumulosus* was the most sensitive species with reference to survival, growth and reproduction. Significant effects on *L. plumulosus* were evident in all sediments treatments except the lowest concentration of cadmium compared to the control. *N. arenaceodentata* showed significant effects only in the two highest concentrations of cadmium for both survival and growth. *M. lateralis* showed significant effects on survival only for the two highest concentrations of cadmium, no significant effects on growth were evident. The differences observed among the species included in this study emphasize the importance of testing sediments using a taxonomically diverse array of bioassays.

PTA232 The SED-TOX Index: a toxicity-directed management tool to assess and rank sediments based on their hazard. Concept and application. Bombardier, M.*, Bermingham, N., Environment Canada, St. Lawrence Centre, Montreal, Quebec, Canada. Several approaches have been proposed to deal with contaminated sediments. Those based on toxicity evaluations have proven particularly useful. The Sediment Toxicity (SED-TOX) Index for the assessment and ranking of (geno)toxic hazard is presented. Major features include: expression of toxicity responses on a single scale of measurement; (ii) consideration of multiple routes of exposure; (iii) application of differential treatments to toxicity data depending on the level of response; (iv) use of weighting factors to discriminate sediment exposure phases and effect endpoints on the basis of sensitivity. A battery of seven bioassays with four test species was conducted on 49 marine sediment samples collected from six sites at Anse-à-Beaufils and Cap-aux-Meules, in the Gulf of St. Lawrence. SED-TOX scores were calculated for each sampling station and compared with sediment contaminant concentrations. Results indicate that physicochemical characterization is not sufficient to assess contaminated sediment hazard for organisms; furthermore, using several exposure phases and test species belonging to various trophic levels increases the possibility of correctly identifying toxic sediments. Based on this study, the SED-TOX approach is valuable as a toxicity assessment and ranking tool for sediments. It could easily be combined with other measures of ecosystem disturbance to discriminate between polluted and unpolluted sites.

PTA233 Relationship Between Microtox™ Extract Toxicity and Sediment Chemical Contamination. B.C. Thompson*, E. Strozier, M. Sanders, L. Reed, S. Sivertsen, M.H. Fulton, and G.I. Scott, NOS, Charleston, SC. Contamination of the estuarine environment may occur due to point source and non-point source pollution. Many pollutants are hydrophobic and adsorb to sediment particles. At sufficient concentrations these contaminants may impact ecosystem health. The bioluminescent bacterium, *Vibrio fischeri*, can be used to rapidly screen sediments for toxicity using the Microtox™ bioassay. The objectives of this study were to determine the spatial distribution of sediment toxicity in South Carolina estuaries and to determine the relationship between Microtox™ toxicity and measured chemical concentrations. Sediment samples were collected from Charleston Harbor, Leadenwah Creek, and Winyah Bay in South Carolina. Chemical analyses were performed using GC/MS. Samples for Microtox™ bioassays were solvent extracted overnight with dichloromethane and exchanged with 5 ml DMSO. EC_{50} values were obtained by measuring the luminescence lost following exposure to the extracts. EC_{50}s from the sites sampled were compared to those from North Inlet, a National Estuarine Research Reserve and Sanctuary. A one-sided Dunnett's test was used to determine sites that were significantly more toxic than the reference site. Cumulative Effects Range Low (ERL) concentrations were then calculated for each site. Cumulative ERL proportions of all non-significant Microtox™ sites were then compared to all sites that were significantly more toxic in the Microtox ™ bioassay ($p<0.05$). A total of 12 sites were significantly more toxic than the reference site, more than 50% of which were in Charleston Harbor. Results of the cumulative ERL proportions indicated that there was a trend toward higher cumulative ERL proportions in the samples that were significantly more toxic in the Microtox ™ bioassay. These findings suggest that Microtox™ toxicity may be indicative of anthropogenic chemical contamination in sediments.

PTA234 Modification of Metal Partitioning by Supplementing Acid Volatile Sulfide in Freshwater Sediments. Leonard, E.N.*, Mount, D.R and Ankley, G.T., U.S, EPA, Duluth, MN. Acid volatile sulfide (AVS) is a component of sediments which complexes some cationic metals and thereby influences the toxicity of these metals to benthic organisms. EPA has proposed AVS as a key normalization phase for the development of sediment quality criteria for metals. Experimental manipulation of AVS in metal-contaminated sediments may provide a means to neutralize toxicity due to metals and thereby help assess the cause of sediment toxicity. This study evaluated the effect of spiking FeS, Na_2S, and $Na_2S/FeSO_4$ combined, on the concentration of AVS, simultaneously extracted metals (SEM), and pore water metals in uncontaminated and metal-enriched sediments. Experiments with solid FeS showed comparatively low effectiveness in increasing AVS. Spiking with either Na_2S, or $Na_2S/FeSO_4$ combined, increased AVS and/or reduced SEM metal in Cd-, Zn- and Ni-spiked sediments, and in a Cu-contaminated sediment collected from the field. Spiking with $Na_2S/FeSO_4$ caused marked reductions in dissolved metal concentrations in the pore waters of these sediments; spiking with Na_2S alone caused an apparent elevation in pore water (Cu) metal, which we believe to be an artifact of metal sulfide formation in the filtered pore water. When the $Na_2S/FeSO_4$ treatment was evaluated under conditions simulating those in sediment toxicity tests, alterations of AVS/SEM were nearly quantitative, except for Ni-spiked sediment which showed lower efficiency than the Cd, Zn, or Cu sediments. It appears that AVS spiking holds promise for the experimental manipulation of metal toxicity in sediments.

PTA235 The Use of Ferrous Chloride to Eliminate Dissolved Sulfide in Sediment Porewater Toxicity Tests. Gorrie, J.R*, Jenkins, C.H., Bidwell, J.R. University of South Australia, Adelaide, S.A. Australia. While sulfides appear to control metal bioavailability in sediments, dissolved sulfide can often occur at toxic levels in sediment porewater making it difficult or impossible to determine effects associated with organic contaminants. We investigated the potential of ferrous chloride additions to remove sulfide toxicity through binding and precipitation of sulfides in porewater. 48-h acute bioassays with the cladoceran *Daphnia caranata*, indicated no effects after moderately hard water (daphnid culture water) was spiked with sulfide ($Na_2S.7H_2O$) to 32 ppm (greater than 100 times the sulfide LC50 for the daphnid), excess ferrous chloride added and the solutions centrifuged to remove precipitates. A phenol solution and the water soluble fraction (WSF) of a crude oil were also spiked with sulfides and the test solutions stripped using the same method. Effects were comparable to that from bioassays with phenol and WSF alone. The 48-h LC50 for the phenol bioassay was 21.9 µg/L (95% Confidence Interval: 18.2-26.4 µg/L) and 19.3 µg/L (15.8-23.6 µg/L) for the phenol-sulfide stripped bioassay. An LC50 could not be calculated for the WSF in either treatment due to low toxicity. This method holds promise for toxicity evaluations of porewater by removing sulfide from test solutions without modifying the action of other contaminants present.

PTA236 The use of a resistant strain of *Chironomus yoshimatsui*, as a positive control for sediment toxicity tests. Sugaya, Y., National Institute for Environmental Studies, Tsukuba, Ibaraki, Japan. A laboratory sediment toxicity test was conducted using both resistant and susceptible strains of the midge, *C. yoshimatsui*, to assess the toxicity of ethopenprox. The bottom mud from Lake Kasumigaura was spiked with ethopenprox, a synthetic pyrethroid insecticide, and prepared a series of mud which contaminated at the concentrations, 0.0 for a negative control, 0.5, 1.6, 7.9 and 58 mg ethfenprox /kg dry mud. The midge larvae of third instar both resistant and susceptive, the sensitivity of those strains to ethofenprox is different over a 1000 folds in 48h-LC_{50} value were reared in laboratory from egg. The test procedure was followed US-EPA guideline, using flow-through aquarium, except turnover rate of overlying water, which was 10 times per a day in the present study, and the exposure time was 7 days, 20 individuals in an aquarium, 4 replicates 4 for both strains. At the highest concentration, the susceptive larvae all died in a 48 hrs, however the mortality of the resistant ones in all concentrations and those of the susceptive larvae at 0.5 and 1.6 mg/kg mud were as low as that of control after the 7 days exposure. At a 7.9 mg/kg mud concentration, about 60 % of the susceptive larvae died. The difference in the mortality of both strains shows that ethopenprox effects on only susceptive strain, and the experimental condition of the present study did not induced any other effects in the mortality, although the other effects, ecological or physical, of chemical were not known yet. The use of a resistant strain in toxicity tests can introduce a good positive control.

PTA237 Utility of a Full Life Cycle Copepod Bioassay Approach for Assessment of Sediment Contaminant Mixtures. C.E. Kovatch*, G.T. Chandler, and B.C. Coull, University of South Carolina, Columbia, SC. Partial life cycle toxicity tests may not effectively measure the most sensitive life stages of an organism. In an attempt to provide a more comprehensive tool for measuring sediment toxicity of mixed contaminants, we compared the utilities of a partial and a full life cycle bioassay of a meiobenthic copepod. We hypothesized that the full life cycle test would better represent chronic exposures to organisms and provide more information on contaminant effects on copepod populations. We compared a 21 day full life cycle bioassay with an existing 14 day partial life cycle bioassay for two species of meiobenthic copepods, *Microarthridion littorale* and *Amphiascus tenuiremis*. *M. littorale* was collected from pristine North Inlet (NI), SC, and *A. tenuiremis* was cultured in the laboratory. Copepods were exposed to sediments from two sites, non-contaminated NI and contaminated Shipyard Creek (SYC), SC, a decommissioned U.S. Naval base. Lethal and sublethal test points included adult survival, clutch size, production of copepodites and nauplii, and potential/realized offspring. Both the 14 and 21 day bioassays showed similar negative effects in SYC sediment for both species. Survival and reproductive success were higher in the 14 day than in the 21 day bioassay for both species in both sediment types. Therefore, the lower 21 day overall response was not indicative of enhanced sensitivity, as it did not provide more information than the 14 day test for reproductive effects. The 14 day bioassay provides sufficient f_1 generation information to detect negative population level responses to contaminant mixtures, and it is more economical to use as an environmental management tool.

PTA238 Combining Sediment Bioassays and Population Genetics to Determine Adaptation of Marine Meiofauna to Contaminated Sediments. C.E. Kovatch*, N.V. Schizas and G.T. Chandler, University of South Carolina, Columbia, SC. The rise of global coastal development necessitates an understanding of subtle, population level effects caused by the concomittant increase in pollution. We hypothesized that meiobenthos which have been chronically exposed to contaminants should exhibit heightened tolerance when compared to meiobenthos which have had little or no previous contaminant exposure. Population level responses to sediment contaminants to the meiobenthic copepod *Microarthridion littorale* were measured by 1) performing 14 day reproductive toxicity tests using field contaminated sediments and 2) estimating genetic diversity of field populations. Copepods were collected from three South Carolina estuaries: North Inlet (NI) a pristine site, Diesel Creek (DC), a USEPA Superfund site, and Shipyard Creek (SYC), a decommissioned U.S. Naval port with pollutant levels higher than DC. Lethal and sublethal toxicity test endpoints included adult survival, average clutch size, production of copepodites and nauplii, and potential/realized offspring production. Results showed that NI *M. littorale* realized offspring production was higher than DC and SYC *M. littorale* production in clean NI sediment. In SYC sediments, the predicted heightened tolerance and offspring production between polluted (DC and SYC) and control (NI) *M. littorale* populations was not realized. Estimated genetic diversity, achieved by analyzing two gene sequences, indicated that there is extensive gene flow among the three populations. Absence of increased tolerance of copepods in contaminated creeks may be explained by the migration of copepods from areas with no contamination history, thus leading to outbreeding depression. The results of the bioassays and the molecular genetic analysis of copepod populations indicate an enhanced reproductive sensitivity to pollutants for *M. littorale* inhabiting contaminated sediments, and not a pollution induced tolerance.

PTA239 Results of the Seed Clam Sediment Bioassay: Comparison of *In situ* Deployments and Laboratory Experiments. Ringwood, A. H., Marine Resources Research Institute, Charleston, SC; Keppler, C.*, Marine Resources Research Institute, Charleston, SC. Sediment toxicity assays were conducted using juvenile *Mercenaria mercenaria* to compare differences in results between laboratory assays and *in situ* deployments. Juvenile clams were deployed for one week at a variety of degraded and undegraded sites in the Charleston Harbor area in February, 1998. Laboratory assays were also conducted using sediments collected from these sites, and growth rates were compared to control site growth. Growth for control sites was similar in both the laboratory and field experiments. At the potentially degraded sites, growth was less *in situ* than in the laboratory. These initial studies suggest that laboratory assays may underestimate potential sediment toxicity at degraded sites. Winter studies will be repeated during the summer to consider seasonal differences in toxicity potential.

PTA240 Evaluation of a Method for Measuring Equilibration Times of Hydrophobic Chemicals with Sediments. Lukasewycz, M.T.*, Mount, D.R., Leonard, E.N., Burkhard, L.P., U.S.EPA, Duluth, MN. Assessment of sediment toxicity often requires the simulation of field conditions in the laboratory. This may include the spiking of highly hydrophobic organic compounds into a test sediment. When spiking high K_{ow} organic compounds into a sediment matrix, it is particularly difficult to quantitatively demonstrate attainment of a steady state partitioning between sediment and pore water, which is critical to simulating chemical behavior similar to that expected in field sediments. A spiking method which delivers a measured, relatively large mass of highly hydrophobic organic compound into a sediment and permits the monitoring of the transfer of the compound into the sediment matrix has been studied. In this method sand particles were coated with a mass of organic compound sufficient to achieve a desired porewater concentration in the test sediment as predicted by the equilibrium partitioning model. The coated sand and test sediment were combined and the mixture was allowed to equilibrate over a period of several months. Aliquots of the test sediment were periodically removed and were fractionated into water, silt and sand fractions using a slurry/rapid settling procedure. Total organic carbon (TOC), particle size distribution and spiked compound concentration were measured in each fraction. Results indicate that the fractionation procedure does effectively separate sand from silt, with the silt fraction containing little or no sand but the majority of the organic carbon, i.e., 72% of the organic carbon. The particle size distribution of the silt fraction was 1.9% for <2 microns, 92% for 2-50 microns and 5.9% for > 50 microns. Redistribution of spiked chemical from the sand fraction to an organic carbon-weighted distribution provides evidence of the attainment of projected equilibrium.

PTA241 Interstitial Water Sampling of Metals in Marine Sediments: Precision of the Dialysis ('Peeper') Method. Serbst, J.R.*, Burgess, R.M., Kuhn, A., Edwards, P.A., Cantwell, M.G., Pelletier, M.C., and Berry, W.J., U.S. EPA, Narragansett, RI. The practice of measuring contaminants in interstitial water (IW) during sediment toxicity tests enables researchers to relate contaminant concentrations to organism responses. There are several methods for collecting IW; here we evaluate the precision of the dialysis or 'peeper' method. In this study, sediment was amended with five concentrations of cadmium and included a control. These sediments were used to conduct seven consecutive ten-day toxicity tests during a seventy day period resulting in a unique data set for assessing method precision. Sediments were equilibrated with flow-through water for seven days before test initiation. IW was sampled on day ten of each toxicity test, cadmium measured, and the data analyzed. Data sets were organized by concentration and test. Precision was expressed as the coefficient of variation (CV) and calculated for each data set. The grand mean CV for all data sets was 77% and CVs ranged from 25 to 206%. This level of variability is comparable to what others have reported for dialysis (e.g., 8 - 133%) but larger than alternative methods (centrifugation, vacuum filtration). We theorize much of the observed variability was the result of artifacts with the dialysis method (incomplete submergence, contamination). A screening procedure was performed to remove these outlier data values. This screen operated as a function of each data set median. Following this procedure and removal of outliers, CVs were ≅30% for all data sets. The dialysis procedure can be an effective means for sampling IW metals; however, care must be taken to avoid method artifacts.

PTA242 Sediment Toxicity Thresholds Based on 90th Percentile MSD Values. B.M. Phillips, B.S. Anderson, and J.W. Hunt, University of California, Santa Cruz, CA. As part of a cooperative program to assess sediment quality in California's bays and estuaries, the California Bay Protection and Toxic Cleanup Program (BPTCP) has used a variety of toxicity test protocols on a total of 1147 sediment samples. When determining the statistical significance of toxicity test results, a number of methods have been employed, including separate variance t-tests comparing samples to laboratory controls. To allow consistency among comparisons, regardless of among-replicate variability, a protocol-specific approach has been used in which Minimum Significant Difference (MSD) values were calculated for each comparison. The value equal to the 90th percentile MSD was then used as a threshold for sample toxicity. This analysis determines the degree of difference from controls that a test is capable of identifying as statistically significant in 90% of cases. 90th percentile MSDs were developed for nine protocols using BPTCP data from as many as 720 stations. Calculations were made using laboratory toxicity data as a percent of the laboratory control. The threshold values as a percentage of the control values were: 75% for *Eohaustorius estuarius* amphipod survival (n=385); 77% for *Rhepoxynius abronius* amphipod survival (n=720); 80% for *Mytilus galloprovincialis* mussel larval development (n=223); 78% for *Strongylocentrotus purpuratus* purple urchin larval development in interstitial water (n=309), 59% for purple urchin larval development at the sediment-water interface (n=109), and 88% for purple urchin fertilization (n=79); 90% for *Haliotis rufescens* red abalone larval development (n=131); 64% for *Neanthes arenaceodentata* polychaete survival, and 44% for polychaete growth (n=335).

PTA243 Distribution of PCBs in Sediments and Benthic Organisms from Korean Coastal Areas. Lee, K.T.*, Seoul National University, Seoul, Korea; Tanabe, S., Ehime University, Matsuyama, Japan; Koh, C.H., Seoul National University, Seoul, Korea. To assess the status of contamination and to elucidate the distribution of polychlorinated biphenyls (PCBs) in Korean benthic environment, PCBs were analyzed in 63 surface sediments and 5 biological samples from Kyeonggi Bay, Namyang Bay, and Lake Shihwa. Based on equilibrium partitioning model, only two sites (K18 and 19) from Kyeonggi Bay showed higher concentration above which adverse biological effects may occur. Some limitations of equilibrium partitioning approach have been discussed. PCBs concentrations in Incheon North Harbor showed distant-dependent decrease in PCB concentrations, indicating the presence of PCBs sources near Incheon North Harbor. The PCB isomer profiles in highest contaminated inner site were similar to those in Kanechlor mixture. The farther sites from Incheon North Harbor showed greater proportions of higher chlorinated biphenyls. The pattern of PCB congeners in a sediment-dwelling sea cucumber, *Protankyra bidentata,* was similar to those in sediment, indicating that this species accumulates PCBs from sediment without biotransformation or selective uptake.

PTA244 Dredging Moss Landing Harbor: Stuck in the Mud. Griffin, D.* and B.R. DeShields, Harding Lawson Associates, Novato, CA; J. Stilwell, Moss Landing Harbor District, Moss Landing, CA. Moss Landing Harbor, located on the Central California Coast at the mouth of Elkhorn Slough off Monterey Bay, serves a variety of uses, from commercial and recreational fishing to recreational boating to marine research. Most areas of the Harbor have not been dredged for 5 years or longer. In recent years, water-borne particles have filled in channels and berth areas in the Harbor, largely due to record rainfalls. Entering/exiting boat traffic currently must be timed around tides, with some boats stranded at their docks and many deep draft vessels unable to navigate the harbor. Approximately 300,000 cubic yards of sediments require dredging to return the Harbor back to full operation. Analyses of sediments from within the Harbor have shown elevated levels of pesticides, mainly DDT and its derivatives. Pesticides are not present because of harbor-related activities, but originate from the Salinas Valley watershed, an extensive agricultural area. Sediment bioassays have shown variable results, with top sediment layers collected in 1997 showing unacceptable levels of toxicity to amphipods. Results from recently collected samples have shown similar levels of pesticides, but lower levels or an absence of toxicity. Another confounding issue is that more cubic yards of sediments, presumably with the same characteristics as those deposited within the Harbor, flow out the mouth of Elkhorn Slough than are deposited in the Harbor and require dredging. To complicate issues further, six state and federal agencies as well numerous County agencies are involved with management decisions regarding Harbor dredging and disposal. An evaluation of the chemical, physical, and biological data for harbor sediments and a discussion of the regulatory process and constraints is presented, along with recommendations for changes, both in the methods of sampling and testing of sediments and in the regulatory framework.

PTA245 Heavy-metal Toxicity in Sediments of Jinhae Bay, Korea. Kong,I.-C., Department of Environmental Engineering, Yeungnam University, Kyungsan city, Kyungbuk Korea. With the increased industrialization of Korea, considerable amounts of chemicals, especially heavy metals, are being introduced into a number of factory districts. Among several of these sites in Korea, Jinhae Bay has recently become one such public concern. In order to meet the demand for rapid screening of toxic pollutants in environments, rapid toxicity tests have to be used. As supported by several investigators, β-galactosidase has shown a high degree of sensitivity to heavy metals while being virtually insensitive to all organic toxicants. Toxicity was correlated ($r^2 = 0.658$) with the total content of Zn, Cd, Pb, Cu, Cr, and Mn. However, a higher correlation ($r^2 = 0.8677$) was obtained between toxicity, and zinc and manganese. Such differences may be due to the binding of heavy metals to solids, and varying toxic effect of metals to enzyme activity. Therefore, determining total content of heavy metals by chemical analyses, is insufficient to assess the environmental impact on contaminated soils and sediments, because it is the chemical forms that determine metal behavior in the environment. Some studies reported little correlation between total heavy metal content and toxicity. This solid-phase toxicity test has several advantages over indirect solid toxicity test. For example, it prevents dilution of toxicants, is specific for heavy metal, and allows a direct contact between contaminants and the test organism or enzyme. Based on these advantages, the test allows a better assessment of the impact of metals on sediments, soils or wastewater sludges.

PTA246 *In Situ* and Laboratory Toxicity Tests with Sediments from Pakuranga Creek, Auckland, New Zealand. Melo, S.L.R.*, EESC -University of São Paulo, Brazil; Morrisey, D.J. and Wise, B.E., NIWA, Hamilton, New Zealand. Toxicity tests were done to evaluate sediment contamination at 3 sites along Pakuranga Creek, a small urban estuary receiving contaminants from stormwater runoff. Two tests were done using interstitial water from the 3 sites: 36-h embryo-development test on fertilised eggs of the sand dollar, *Fellaster zelandiae* and 96-h water-only test on the amphipod, *Chaetocorophium lucasi.* Interstitial water toxicity was observed in both sand dollar and amphipod tests for all sites. *C. lucasi* were also exposed to sediments from the 3 sites in both the laboratory and the field. Survival was measured over 10 days (acute test) and survival and growth over 28 days (chronic test). *In situ* cages, containing a 2-cm deep layer of sediment from one of the sites and extra cages containing control sediment collected from a pristine estuary were used. Amphipod survival in laboratory control sediment was 93.3% and 85% for acute and chronic tests, respectively. In the acute test, amphipods exposed to sediment from one of the 3 contaminated sites showed reduced survival relative to controls. In the chronic test, animals on the control sediment grew faster than those on sediments from all contaminated sites. In the *in situ* tests, amphipod survival in the control was 85% in average. Survival in the contaminated treatments was not different to the controls. Amphipods had grown significantly after 28 days at all sites. Amphipods in control cages deployed close to the estuary's mouth showed more growth than in controls deployed higher up the estuary and in the remaining treatments, suggesting an effect of external environmental factors in addition to the effect of the sediments from different sites.

PTA247 Effects of Dredge-Spoil Runoff in the Savannah River. Winger, P.V.* and Lasier, P.J., USGS Patuxent Wildlife Research Center, Athens, GA. The lower Savannah River distributary, consisting of Front, Middle and Back rivers, encompasses the city of Savannah, GA, Savannah Harbor and the Savannah National Wildlife Refuge. Harbor activities (maintenance and expansion), in conjunction with municipal and industrial discharges, have adversely affected habitat quality and indigenous fish and wildlife resources. The objectives of this study were to ascertain the effects of dredge-spoil runoff on habitat quality in the river. Sediments from 35 river sites and 6 dredge-disposal sites were collected for analysis of metal and organic contaminants (organochlorine and organophosphate pesticides, PAHs and PCBs) and toxicity testing of solid-phase sediment and sediment pore-water using *Hyalella azteca*. Bioaccumulation of contaminants from 6 dredge-spoil sediments was determined using *Lumbriculus variegatus.* Metal residues were measured in livers of marsh ducks (*Anas creca*), wading birds (*Tringa flavipes*) and raccoons (*Procyon lotor*). Most sediments were not toxic, and for those showing toxicity, water quality characteristics (ammonia, alkalinity, salinity) were generally responsible. However, dredge-spoil runoff or point-source discharge may have contributed to the toxicity shown at 14 sites. Organic contaminants in sediments were at or below detection limits. *Lumbriculus* bioaccumulated arsenic, copper, mercury, selenium and zinc from dredge-spoil sediments. Cadmium, mercury, molybdenum and selenium residues were higher in livers of birds and raccoons than in sediment, and these metals were significantly higher in livers of raccoons living near the river than those living in an upland control area. Evidence of bioaccumulation from laboratory and field evaluations, toxicity of sediments from some areas receiving dredge-spoil runoff and concentrations in sediments demonstrated that some metals present in the dredge spoils are mobile and biologically available and have the potential to impact habitat quality in the lower Savannah River.

PTA248 Development of an *In Situ* Model for the Toxicological Evaluation of Sediments in Large River Systems. Blanner, P. M.*, and Maier, K. J., The University of Memphis, Memphis TN. *In Situ* testing methods have been proven to be an effective tool for the toxicological evaluation of sediments. While numerous studies have demonstrated the efficiency of *in situ* techniques, there exists a need for the development of *in situ* testing methods for use in large river systems. This study addresses this issue and proposes an *in situ* model for the toxicological evaluation of sediments in large river systems. The study was conducted in McKellar Lake, an inlet of the Mississippi River at mile 725.6. *In situ* exposure chambers were designed to allow for maximum water flow and sediment contact. The test organisms used were *Chironomus tentans* and *Hyallela azteca*. The organisms were exposed to McKellar Lake sediments both *in situ* and under laboratory conditions for a period of seven days. Preliminary experiments have demonstrated greater than eighty percent survival of control organisms in both laboratory and *in situ* conditions, thus suggesting that this *in situ* chamber design is suitable for use in large river systems.

PTA249 Toxicity Assessment of Historically Contaminated Sediments. Huggett, D.B., Muller S.L., Bohannon, A.L., University of Mississippi, Oxford, MS; Rodgers, J.H., Clemson University, Pendleton, SC; Adams, D.L., International Paper, Memphis, TN; Deardorff, T.L., International Paper, Cincinnati,OH. To learn more about remediation options for historically contaminated lake sediments, we collected samples from various locations within a southeastern reservoir that previously received discharges from several facilities. Our initial assessment showed that although most of the sediment samples were not problematic, some material(s) in a few sediment samples adversely affected *Chironomus tentans* survival and growth, *Ceriodaphnia dubia* survival and reproduction, *Hyalella azteca* survival, *Pimephales promelas* survival and *Typha latifolia* seed germination and early seedling growth. To better estimate the degree of toxicity, sediment dilution experiments (definitive sediment toxicity tests) revealed that as much as 87.5% dilution was required to decrease observed toxicity to reference levels. Laboratory remediation assessments with sediments from two locations in the reservoir focused on oxidation (either through aeration or H_2O_2 treatment), addition of activated charcoal and *in situ* capping. Oxidation of both sediments, either through aeration or H_2O_2 treatment, decreased observed toxicity to the sentinel testing organisms. Addition of activated charcoal decreased observed toxicity at one site; however, toxicity in the other sediment was essentially unaffected by this treatment. Experimental capping of both sediments illustrated that a 1.25 cm sand cap was adequate to decrease all observed toxicity exhibited by the sentinel organisms. Since these laboratory experiments may not translate directly to the field, a longer oxidation time and a deeper cap may be required in the field to obtain the same mitigation of toxicity observed in the laboratory.

PTA250 Mercury Contaminated Sediments - Evaluation of Environmental and Human Health Risks. Duh, D.J.*, IT Corporation, Somerset, NJ; Medeiros, W.H., Brookhaven National Laboratory, Upton, NY; Ali, M., DOE, Upton, NY; Watt, M., IT Corporation, Somerset, NJ; Luke, N., IT Corporation, Somerset, NJ. A remedial investigation discovered contamination in the sediments of an intermittent headwater stream of a larger river. Several metals, pesticides, and PCBs with bioaccumulation potential were found in these sediments. The highest concentrations were found in depositional areas located on site; decreasing concentrations of some contaminants were found off site. Toxicity tests indicated that the higher concentrations of metals, particularly mercury, in the depositional areas on site presented a risk to benthic invertebrates. Additionally, concentrations of mercury and fish captured on-site posed a potential risk to piscivorous wildlife. The concern for human health protection led to an intensive collection of fish both on site and off site; off-site collections included "non-fishable" areas near the site and "fishable" areas further off-site. No current risk to human health was found. However, the concentrations of PCBs in fish found on-site could pose a risk to humans whose fish consumption consists entirely of on-site fish in the absence of current institutional controls. Remediation of contaminated sediments located on site is currently being addressed.

PTA251 Effects of Biological and Procedural Factors on Reproductive Output in the *Chironomus tentans* Life Cycle Test. Sibley, P.K.*, Centre for Toxicology, University of Guelph, Guelph, Ontario; Ankley, G.T., USEPA, Mid-Continent Ecology Division, Duluth, MN. A life cycle test using the midge *C. tentans* was recently introduced for assessing chronic toxicity of contaminated sediments. In this test, factors affecting traditional endpoints (survival, growth) have been relatively well documented based on experience with the *C. tentans* 10-d test. In contrast, factors affecting reproductive endpoints (e.g., # eggs/female, percent hatch) in the life cycle test are poorly understood. The objective of this study, therefore, was to assess the relative influence of several physiological and procedural factors on reproductive output of *C. tentans*, including: i) time to emergence, ii) weight of adults, iii) time to first mating for ♂ and ♀, and iv) number of matings for ♂. A life cycle test was conducted using a clean natural sediment. Sixteen replicate beakers were placed in each of six treatments, defined as the number of days (1-6) that ♂ or ♀ were delayed from mating. Reproductive output increased significantly with an increase in ash-free dry weight (afdw) of females, but not males. Both ♂ and ♀ afdw increased with increasing time to emergence. Males successfully mated an average of 4.5 times, with no temporal change in the weight-adjusted reproductive output of females, when mating occurred on consecutive days. However, if males mated more than once in 24 hr, or mating frequency increased to 6-7 events, reproductive output declined significantly. Time to first mating (delay) was not significantly related to reproductive output in either ♂ or ♀. While these data support the use of a single male for multiple mating events, the time-dependent increase in reproductive output of females may be important to consider when interpreting reproductive toxicity in contaminated sediments using the life cycle test.

PTA252 Activities to Understand the Risk of Surfactants in Sediments: Long Term Efforts of The Soap and Detergent Association. DeCarvalho, A.J., The Soap and Detergent Association, New York, NY; Dyer, S.D. The Procter & Gamble Co., Cincinnati, OH; Cano, M.L., Shell Development Co., Houston, TX. Over 5 million metric tons of surfactants are consumed worldwide, primarily in laundry and cleaning products. A residual amount is discharged to the environment after consumer use and removal via wastewater treatment. These surface-active compounds have a propensity to sorb to suspended particulates and sediment. To understand the relationships of surfactant sorption to solids, bioavailability, toxicity and ambient sediment concentrations, the Soap and Detergent Association has maintained a long term program devoted to addressing each of these key areas. Sorption experiments were conducted using linear alkylbenzene sulfonates (LAS) and alcohol ethoxylates (AE). The experiments showed that the sorption of LAS with three different alkyl chainlengths (C10, C12 and C14) was dependent on the fraction of organic carbon (foc) in the sediment. pH and ionic strength (Ca2+) of the test solution were also important in LAS-sediment sorption. In contrast, AE-sediment sorption experiments with 3 different ethoxylate chainlengths (C13EO3, C13EO6, C13EO9) indicated that pH, ionic strength and foc were not correlated with the extent of sorption. The bioavailability of a model surfactant (highly branched alkylbenzene sulfonate (ABS)) was determined via acute sediment toxicity tests with the amphipod *Hyallela azteca*. Results showed that sorption, hence bioavailability, was dependent on foc. Toxicity in sediment was due to interstitial water exposure. Progress on extraction and analytical methodology for several classes of surfactants to determine ambient concentrations on sediments will also be presented.

PWA001 Effect of Methyl-tert-butyl ether (MTBE) on the phototoxicity of polycyclic aromatic hydrocarbons (PAH). Choi, J.*, Oris, J.T. Miami University, Oxford. OH. 45056. The 1990 Clean Air Act Amendments require fuel oxygenates to be added to gasoline used in some metropolitan areas to reduce atmospheric concentrations of carbon monoxide and ozone. To meet the requirement gasoline must contain at least 15% MTBE by volume. Even though oxygenates are currently used in more than 30% of the US gasoline pool and this figure is expected to reach as much as 70% by the end of the century, few studies have been conducted on the effect of MTBE on aquatic ecosystems. MTBE enters aquatic systems through accidental gasoline spills, urban runoff and emissions from motorized watercraft. MTBE and PAH commonly co-occur in aquatic systems near urbanized areas and areas with motorized watercraft activity. It has been well established that PAH are acutely toxic to a wide range of aquatic organisms below water solubility limits in the presence of solar ultraviolet radiation (SUVR) of environmentally realistic intensities. In this study, the possibility of MTBE potentiation of PAH phototoxicity was investigated using fathead minnow larvae. Fathead minnow (*Pimephales promelas*) larvae were simultaneously exposed to a model PAH (20μg/L of fluoranthene) and simulated sunlight (UV-A of $74 \mu W/cm^2$) with or without 1μg/L of MTBE. This level of MTBE concentration was not toxic to larvae as itself. The addition of

MTBE was found to increase the mortality of fathead minnow larvae in these studies indicating a potential synergistic reaction between combustion by-products and gasoline additives.

PWA002 **Survival and Teratological Effects of Copper on Embryo-Larvae of the Inland Silversides,** *Menidia beiyllina.* Barber, R.D.*, University of Houston - Clear Lake, Houston, TX; Hollister, T.A., U.S. EPA, Houston, TX. Current short-term chronic, seven-day test utilizing 7-11 day-old inland silverside, *Menidia beryllina,* larvae are labor and time intensive, and have been known to provide significant variation (30%) in the 1C50's for survival and growth. A more efficient and slightly more sensitive seven-day test was developed utilizing *M. beryllina* embryo-larvae spawned in the laboratory. Six tests were executed in which 84±12 hour-old embryos were exposed for 7-days to 5 test concentrations of copper in HW Marinemix® artificial seawater. The mean 1C25 and 1C50 for the tests were 157 and 195 ppm, respectively. Corresponding coefficients of variation were 4.0% and 6.3%. In using *Menidia* embryos, only 11 days were required to prepare and execute a test versus the minimum of 22 days required by the EPA approved method using *Menidia* larvae. The time savings, excellent repeatability, overall sensitivity, and simplicity of execution, suggests the proposed method be evaluated further and that it be investigated as a possible alternative to the current methods used for estimating short-term chronic toxicity.

PWA003 **Dietary selenium enhances the elimination of methyl mercury by shrimp** *Crangon crangon.* Hunter, D.A.*, Christensen, A., Odense University, Odense, Denmark. Exposure of mercury-contaminated fish to selenium in water and especially food is known to enhance the elimination of mercury. In the present study, shrimps (*Crangon crangon*) contaminated with methyl mercury were fed selenium (as selenite) to determine whether ingested selenium increases the rate of elimination of mercury by crustaceans. Five days after consuming food containing radiolabelled methyl mercury ($CH_3{}^{203}HgCl$) shrimps were fed (at 2% of body weight d^{-1}) meals dosed with 10 or 20 µg selenium (as selenite) g^{-1} wet weight, or control food, daily for 17 days. Hereafter, half the shrimps from each group were dissected to determine mercury distribution and selenium concentrations in tissues, while the remainder were fed undosed food for 5 days. An immediate and significant, dose-dependent increase in the rate of elimination of mercury was seen in selenium-fed shrimps. Half-lives for methyl mercury were ~ 100 and 50 days, respectively, in shrimps fed 10 or 20 µg selenium g^{-1} wet weight food. Control animals showed no measurable loss of mercury (methyl mercury half-life in the crab, *Carcinus maenas,* is ~ 750 days). The selenium-fed animals showed no further loss of mercury when given control food. Analysis of muscle, gill, midgut, exoskeleton, rest, and other (fluids lost on dissection) tissues showed the rest and other 'tissues' of selenium-fed shrimps to contain, respectively, relatively less and more of the whole body ^{203}Hg activity. Muscle selenium concentrations did not increase significantly. These results demonstrate that ingested selenium can augment elimination of mercury by crustaceans as it does in fish. Feeding fish diets containing elevated, but still low, levels of selenium in order to reduce mercury burdens to levels considered fit for human consumption may also be a strategy applicable to certain wild-caught and farmed crustaceans.

PWA004 **Importance of Calcium on Zn Accumulation on Fish Gills.** Welsh, P., Lipton, J., Barron, M., Hagler Bailly Services, Inc., Boulder, CO.; Playle, R., Wilfrid Laurier University, Waterloo, ONT. Different concentrations of calcium in acclimation and exposure water have been correlated with differential uptake and accumulation of Zn on rainbow trout gills. Zinc accumulation was lowest when fish were acclimated in hard water (Ca = 120 mg/L) and tested in hard water and highest when fish were acclimated in soft water (Ca = 8 mg/L) and tested in soft water. Intermediate Zn accumulation was observed when fish were either acclimated in soft and tested in hard water or acclimated in soft and tested in hard water. Total Zn concentrations in gills ranged from 20 to 55 μg/kg fish in the different treatments after 24 hours exposure to 100 μg ^{65}Zn/L. Calcium can affect Zn accumulation by competition reactions with binding sites on the gill surface and by affecting gill ion membrane permeability. Using gill ligand constants for Ca and Zn, we modeled the gill-Zn concentration on the gill surface over time in the various exposure waters tested using the geochemical speciation program MINEQL$^+$. The relative importance of the role of Ca in competition versus acclimation in Zn accumulation on fish gills will be discussed

PWA005 **Calcium-Dependent Zinc Uptake by Rainbow Trout.** Barron, M.G.,* Albeke, S., Hagler Bailly Services, Boulder, CO. Calcium is known to control the uptake and toxicity of zinc and other metals through chemical competition, biological acclimation, or both. The dominant process controlling zinc uptake has not been previously elucidated. We determined zinc uptake by rainbow trout acclimated and exposed to four calcium treatments: (1) low calcium (30 mg Ca/L) acclimation and low calcium exposure (LL); (2) low acclimation and high calcium (120 mg Ca/L) exposure (LH); (3) high acclimation and low exposure (LH); and (4) high acclimation and high exposure (HH). Trout were exposed to sublethal zinc (100 μg ^{65}zinc/L) for 24 hours, and whole body and gill ^{65}zinc levels were determined. Zinc uptake was approximately linear during the 24 hour exposure period and uptake was calcium dependent. Zinc uptake by trout was 22 μg/kg·hr in the LL treatment and 4.5 μg/kg·hr in the HH treatment. Zinc uptake by trout in the LH and HL treatments was 13 and 10 μg/kg·hr, respectively. The results of this study demonstrate that calcium reduces zinc uptake through both chemical competition and biological acclimation, and that calcium effects are approximately additive.

PWA006 **Trace Zn accumulation in both soft tissue and shell of the abalone** *Haliotis divesicolor supertexta* **through the alga** *Gracilaria tenuistipitata* **var.** *liui.* Lin, M.-C.*; Liao, C.-M., Department of Agricultural Engineering, National Taiwan University, Taipei, Taiwan, ROC. The accumulation of zinc in the abalone *Haliotis divesicolor supertexta* and the red alga *Gracilaria tenuistipitata* var. *liui* was measured in order to estimate the bioaccumulation of zinc and to indicate the concentration of zinc in the aquatic ecosystems. *G. tenuistipitata* var. *liui* is the major forage for culturing the abalone *H. diversicolor supertexta.* These two aquacultural species are considered to be important aquatic products in Taiwan. The coastal regions where the aquacultures of the alga and abalone are located, however, are subjected to polluted discharges of zinc from rivers. The present investigation originated as an effort to analyze the two species for assessing the bioconcentration and biomagnification in an aquacultural system. Living thalli of the alga were exposed to varied concentrations of zinc, and then fed to the abalone. The results showed that the uptakes of zinc accumulation in both soft tissue and shell of *H. diversicolor supertexta* increased linearly with the amount of zinc in *G. tenuistipitata* var. *liui* which is proportional to the zinc concentration in the sea water. The values of the bioconcentration factor in the algae and the biomagnification factor in the abalone were 625 and 1.85 respectively. These two species are potentially useful biomonitoring systems for the bioaccumulation of pollutants in the artificial and spontaneous environments. The alga accumulated great amount of zinc at a wide range of salinity ranging from 15 to 35 o/oo, therefore it is an ideal material to remove heavy metals. The measurement of heavy metals in the shell of abalone can be utilized as an inexpensive indicator for monitoring polluted abalone, algae or seawater.

PWA007 **Effects of Methylmercury Exposure on a Model Aquatic Predator-Prey System.** Carter, M.K. and Benton, M.J.*, East Tennessee State University, Johnson City, TN. The effect of methylmercury exposure on largemouth bass (*Micropterus salmoides*) predatory behavior was tested in microcosms and mesocosms. In microcosm experiments, a single bass, either mercury-exposed (5 or 10 µg/L) or unexposed, was acclimated in a 38-liter tank for 48 hours. Three fathead minnows (*Pimephales promelas*), either mercury-exposed (5 or 10 µg/L, 6 h) or unexposed, then were introduced into the tank, and time to each capture was recorded. In a second similar experiment, 10-cm-square refugia were affixed to tank bottoms. Times to captures 1, 2 and 3 were significantly affected by refugium presence; times to captures 2 and 3 were significantly affected by predator and prey exposure concentration. Exposed bass failed to capture prey more frequently than unexposed bass. In mesocosm experiments, a single mercury-exposed or unexposed bass was acclimated for 24 hours in a 2-meter-diameter circular tank containing a variety of artificial structure. Twenty mercury-exposed and 20 unexposed fathead minnows (identified by cold brands) then were introduced into the mesocosm, and the bass was allowed to feed for 12 hours. Unconsumed prey then were retrieved and counted, and the remainder were assumed to have been consumed. Unexposed bass consumed significantly more exposed prey than unexposed prey. Exposed bass also consumed more exposed than unexposed prey, but the difference was not statistically significant.

PWA008 Genetic and Demographic Responses of Mosquitofish (*Gambusia holbrooki*) Populations Exposed to Mercury for Multiple Generations. Tatara, C.P.*, Savannah River Ecology Laboratory, Aiken, SC; Mulvey, M., Savannah River Ecology Laboratory, Aiken, SC; Newman, M.C., Virginia Institute of Marine Science, Gloucester Point, VA. Genetic and demographic responses of mosquitofish exposed to mercury for multiple generations were examined in this study. Our previous studies of acute lethal exposures of mosquitofish to either mercury or arsenic demonstrated a consistent correlation between time-to-death and genotype at the glucosephosphate isomerase (*Gpi-2*) locus. A mesocosm study involving mosquitofish populations exposed to mercury for 111 days showed significant female sexual selection and fecundity selection at the *Gpi-2* locus. Here, the mesocosm study has been extended to populations exposed to mercury for multiple (circa 4) generations. Eight populations (4 control and 4 mercury-exposed) were established with fish from an uncontaminated site. After two years, all populations met Hardy-Weinberg expectations and showed no evidence of genetic bottlenecks. Average heterozygosity (calculated with 7 polymorphic loci) did not differ significantly between the mercury-exposed and control populations. Analysis of selection coefficients showed significant differences between the mercury-exposed and control populations at the *Gpi-2* locus. The $Gpi\text{-}2^{100}$ allele frequency decreased (p = 0.055), the $Gpi\text{-}2^{66}$ allele frequency increased (p=0.056), but the $Gpi\text{-}2^{38}$ allele frequency did not change in the mercury-exposed populations. There were no significant differences in average standard length, average weight, sex ratio, or age class ratio between the control and mercury exposed populations. Allele frequency changes at the *Gpi-2* locus suggest population-level response to chronic mercury exposure. Numerous reports of relationships between *Gpi-2* allozymes and population or individual response to toxicant exposure suggest that such changes may be useful as indicators of population response to contaminants, provided that the population in question is well understood.

PWA009 Influence of Body Size on Trace Metal Concentrations in the Insect *Chaoborus*. Alfaro, C.*, Hare, L., Tessier, A. INRS-Eau, Sainte-Foy, Québec, Canada. Trace metal concentrations in aquatic animals tend to vary with body size. Relationships between trace metal concentrations and organisms body size can be grouped into 3 general categories: a) metal concentrations decrease with increasing body size; b) metal concentrations and body size are independent; c) metal concentrations increase with increasing body size. We determined which of these categories best describes the body weight / metal concentration relationship for 4 metals (Cd, Cu, Pb and Zn) in 2 species of the aquatic insect *Chaoborus*. To measure metal concentrations in these small larvae (75-1,030 µg dry weight) we tested and perfected techniques to permit the digestion and analysis of small tissue samples. We used a room temperature digestion (100 µL HNO_3 : 40 µL H_2O_2 for 5 d) and we measured metals by Zeeman Correction Graphite Furnace AAS. For a given larval stage (4th instar), the concentrations of Cu and Zn were independent of larval size for 1 of the 2 *Chaoborus* species. Concentrations of Cd, Pb, and Zn (for one of the two species) decreased with increasing body size, that is, metal in animals was diluted by body tissues added during growth.

PWA010 Metal Accumulation and Acute Toxicity in Brown Trout Exposed Chronically to a Mixture of Cadmium and Zinc. L.L. Gasser, H.S. Ramsdell, Center for Environmental Toxicology and Technology, Department of Environmental Health, Colorado State University, Ft. Collins, CO, S.F. Brinkman, P.H. Davies and J.D. Woodling, Colorado Division of Wildlife, Ft. Collins, CO. In common exposure scenarios in Rocky Mountain streams impacted by mining activity, fish are exposed to mixtures of metal contaminants and intermittent pulses of higher metal concentrations. This study was intended to examine the effects of chronic exposure of fish to a mixture containing low levels Zn and Cd on the acute toxicity of the two metals at higher concentrations and the patterns of metal accumulation associated with acclimation. Juvenile brown trout (*Salmo trutta*) were exposed to sublethal concentrations of Cd and Zn for 180 days. Acute LC50 challenge assays were performed using a Cd/Zn mixture after 90 and 180 days of exposure. Other individuals from the chronic exposure groups were sampled for analysis of metals and metallothionein (MT) content in liver and kidney. The LC50 value was increased by over 2-fold after 180 days of exposure to 0.5 ug/L Cd and 200 ug/L Zn. An approximately 3-fold increase in LC50 was noted in fish exposed at twice these levels. Total Cd concentrations in both liver and kidney increased to a greater extent than Zn levels. Total MT concentrations were 5- and 3- fold higher in liver and kidney, respectively, in fish exposed to 1 ug/L Cd and 400 ug/L Zn but no increases in total MT were observed in fish treated chronically at the lower dose. These results suggest that metallothionein induction alone is not responsible for the acclimation effect observed.

PWA011 Toxicity of Lead, Copper and Zinc Mixtures to *Ceriodaphnia dubia* and *Daphnia carinata*. Bidwell, J.R.*, Kumar, A., School of Pharmacy and Medical Sciences, University of South Australia, Adelaide, Australia. Acute and chronic bioassays were conducted to determine effects of binary and tertiary mixtures of lead (Pb), copper (Cu) and zinc (Zn) upon two species of cladoceran, *Ceriodaphnia dubia* and *Daphnia carinata*. Levels of metals used in the tests were based on current Australian water quality guidelines for single-chemicals (5 µg/L for Cu and Pb and 50 µg/L for Zn), with moderately hard water (80-100 mg/L as $CaCO_3$) as the diluent. In Australia, these criteria are often used to set effluent discharge limits and have also been used to evaluate the quality of urban stormwater. Copper, lead and zinc combined at criterion concentrations did not cause significant mortality of the daphnids during acute exposures, although tertiary mixtures significantly reduced the reproductive output of both species. In *Ceriodaphnia*, the binary combinations of Cu + Zn and Cu + Pb had a significant effect on reproduction at one quarter of the respective criterion levels, while Pb + Zn had a significant effect at half the criterion levels. The joint action of the metals was either additive or more than additive based on toxic units calculated from the acute and chronic tests. The data indicate that single chemical criteria alone may not provide adequate permit limits for the release complex effluents. Focus on single chemical concentrations may similarly underestimate the toxicity of other complex mixtures such as urban stormwater.

PWA012 Toxicity of the 13 priority metal pollutants to *Vibrio fischeri* in the Chronic Microtox Test. Hsieh, C.Y.*, Ryan, D.K., University of Massachusetts-Lowell, Lowell, MA; Pancorbo, O.C., Massachusetts Department of Environmental Protection, Lawrence, MA. The Microtox Acute Toxicity Test has been successfully used to measure the toxicity of metals and other pollutants at high concentrations (ppm) in selected environmental samples. However, metals and other toxicants are often found in much lower concentrations (ppb) in many municipal wastewaters and receiving waters. In order to assess the toxicity of these pollutants in these samples, a more sensitive toxicity assay is needed. Chronic Microtox Toxicity Test has been recently developed by Azur Environmental (Carlsbad, CA) that measures the sublethal effect of toxicants over multiple generations of the test species, *Vibrio fischeri*. In this study, the toxicity of the 13 priority metals [i.e., As, Se, Cd, Cr (III), Cr(VI), Cu, Pb, Sb, Ag, Tl, Zn, Be, Hg and Ni] to *Vibrio fischeri* was evaluated using the Microtox Chronic Toxicity Test. In this test, the Inhibitory Concentrations (IC), Lowest Observable Effect Concentration (LOEC), and No Observable Effect Concentration (NOEC) were obtained after 22-h of incubation at 27±1 °C, by comparing the light output of the control to that of the test sample. Among the 13 priority metal pollutants, beryllium (Be) was found to be the most toxic in the test (LOEC=1.5 µg/L) while thallium (Tl) was the least toxic (LOEC= 5000 µg/L). The LOECs for copper (Cu) and lead (Pb) in reagent (ASTM Type I) water were 20 µg/L and 1,000 µg/L, respectively. The toxicity of Cu (as $CuSO_4$) in reagent water was shown to be significantly reduced with the addition of natural organic matter (NOM) or EDTA to the sample. The LOEC values for the 13 priority metal pollutants in this test were comparable to those reported for commonly used aquatic toxicity tests, such as the *Ceriodaphnia dubia*.

PWA013 The Use Of Periphyton To Determine The Influence Of Heavy Metal Laden Waters In The Powell River. Yeager, J. L.*, Campbell, A.J., Cherry, D.S., Bidwell, J.R., and Zipper, C. Virginia Tech, Blacksburg, Virginia. A periphyton study was conducted to determine if the heavy metal laden waters of Black Creek located in Wise county, Virginia would influence the biota in the main stem of the Powell River. Six sites were selected, one above the creek discharge size (5.0 million gal/day) which was used as a reference, one within and 4 below at 7m, 17m, 200 m and 300 m from the discharge. Clay tiles glued to bricks were placed in the creek and river for 21 days and then returned to the lab. Each brick contained 8 tiles with 4 bricks per site. The tiles were scrapped and the material used to determine Chloropyll a content, ash free dry weight, species diversity and cell count. Chloropyll a amounts were significantly greater at the reference than at the creek site and the two closest Powell River sites with a ten-fold difference between the reference and creek site. There were no differences in ash free dry weight at any of the sites nor were there any differences in species diversity (replicates ranged from 7 to 9 species identified to genus). There were differences in the # of cells/ml at the reference and the creek site as well as the first two Powell River sites with the reference containing a minimum of 36% more cells. This indicated that the creek water was influencing the total number of species present but after 17m the river was no longer influenced.

PWA014 The Effects Of Acid Mine Drainage On Benthic Macroinvertebrate Communities And Leaf Decomposition In A Small Watershed In Southwestern Virginia. Yeager, J. L.*, Campbell, A. J., Cherry, D.S. and Bidwell, J. R., Virginia Tech, Blacksburg, Virginia. A leaf pack study was conducted in conjunction with a benthic macroinvertebrate study to determine the effects of acid mine drainage, in the Black Creek watershed, Wise County, Virginia, on leaf decomposition. Eight sites with pH that ranged from 2.76 to 7.68 were selected as well as one reference site. At each site .5 mm pecan bags with 5.0 grams of *Acer sacchrin* (Sugar Maple) leaves inside were placed in series in the stream. Each site had contained four replicates of four and samples were removed at 24 hrs to determine the leaching rate and again on day 7, 21, and 35. Samples were returned to the lab and measured for penetrance and dry weight. Comparisons were made based on the rates obtained at the reference site. These results were then compared to the findings of an extensive macroinvertebrate survey, with seven samplings over 18 months, two using RBP and the remainder using a quantitative method. Dry weight and penetrance of leaves both reflected slight differences in the decomposition rate after seven days when comparing the reference to the sites with a pH below 5. After 21 days these differences became significant and after 35 days, sites that had been moderately impaired (pH of 5 to 7, high metals) became significant. The macroinvertebrate data reflected this trend, with no insects in streams with low pH below 5, and only *Corydalus* and Hydrophychidae species in moderately impacted areas. Sites that did not differ from the reference contained similar benthic communities composed of multiple species(14-18 taxa, 100+ organisms/replicate) including shredders.

PWA015 Relative Toxicity of Acid-Mine Drainage Water Column and Sediments to *Daphnia magna*. Soucek, D.J.*, Cherry, D.S., Virginia Polytechnic Institute and State University, Blacksburg, VA. Laboratory tests were conducted with *Daphnia magna* to compare the toxicity of acid-mine drainage (AMD) water column and sediments from a Virginia stream. *Daphnia magna* was selected as the test organism because of its activity in both the water column and sediment surface, where it disturbs particles for filter feeding. Water and sediment samples were collected from ten stations in a stream with two AMD point sources and its receiving stream, which is part of the Powell River watershed in Southwestern Virginia. Water column pH values ranged from near neutral at reference stations to 3.12 at point sources, while conductivity values ranged from less than 200 to 2,000 µmhos/cm at reference stations and impacted stations, respectively. Acute (48-h) and chronic (7-d) water column and sediment toxicity bioassays were conducted in the laboratory according to standard methods. For acute sediment bioassays, serial dilutions were made with formulated sediment so that LC50 values could be calculated. Water and sediment samples were analyzed for iron, copper, zinc, aluminum, and manganese. Water column toxicity was observed at several stations with 48-h LC50 values of less than 2.5% at point sources and 14% one mile downstream. Sediment samples also were toxic to *D. magna*, with 48-h LC50 values as low as 58 percent for a station near one of the point sources; however, daphnids were less sensitive to contaminated sediments than to AMD water samples. Based on correlation analysis with benthic macroinvertebrate community data for the ten stations, water column toxicity was a better predictor of *in situ* effects than sediment toxicity.

PWA016 Evaluation of Mine Drainage in USEPA, Region 8 Using Water Column Toxicity Tests, Sediment Toxicity Tests, and A Benthic Macroinvertebrate Indicator. [1]Klemm, D.J.; [2]Herlihy, A.; [1]Lazorchak J.M.;[3] Smith, M.E. and [3]Thoeny. W.T. [1]U.S. EPA, NERL, Cincinnati, OH; Oregon State University, c/o U.S. EPA, Corvallis, OR; [3]SBI Environmental, Inc., c/o U.S. EPA, Cincinnati, OH. The Southern Rockies Ecoregion contains almost 95% of the mineralized portion of the Rocky mountains. For the past century, extensive mining of metals has occurred in this area. Runoff and drainage from both active and inactive mining sites have contaminated waters and sediments. In 1994 and 1995, the U.S. EPA conducted a probability survey as part of its Regional Environmental Monitoring and Assessment Program (REMAP) in the Colorado portion of the Southern Rockies. The survey targeted second-fourth order streams as represented on USGS 1:100,000 scale maps. Using data from this study, the potential impact of mining on stream condition was assessed by three approaches. First, chemistry data collected from the survey was used to classify streams based on ANC, SO_4^{-3} and Cl^- concentrations of the water. High sulfate and metal concentrations in these sites serves as an excellent indicator of mine drainage impacts in the watershed. In the second assessment approach water column samples and sediment samples were collected at the same sites as the chemistry samples. *Ceriodaphnia* and fathead minnow 48-hr tests were conducted on the water column samples and 7-day survival and growth tests using *Hyalella azteca* were performed on the sediment samples. The third approach used stream macroinvertebrate assemblages collected at the sample site to quantify the EMAP multimetric Stream Benthos Integrity Index (SBII) method. The results indicated that almost all of the sites with impacted benthos and water column toxicity occurred in sites classified as mine drainage impacted by water chemistry. The stream chemistry data estimated that 28% of the stream miles in the target population were impacted by mine drainage. The toxicity data estimated that 9% of the target stream length had stream water toxic to aquatic organisms. The SBII scores indicated that 8% of the target stream length had impaired benthic biotic integrity.

PWA017 Toxicity and Bioavailability of Chelated-Copper Herbicides Clearigate®, Cutrine® -Plus, and Copper Sulfate to Freshwater Organisms. Mastin, B.J.*, University of Mississippi, University, MS; Rodgers, J.H., Clemson University, Pendleton, SC. The short-term effects and bioavailability of chelated-copper herbicides in terms of survival of four freshwater testing organisms were assessed in a series of aqueous experiments. *In vitro* toxicitv data contrasting responses of nontarget freshwater organisms to aquatic herbicides are important for understanding sensitive species relationships (exposure-response slopes), development of baseline point estimates of toxicity (LC5Os, LOECs), and *in situ* extrapolation. Aqueous 48-h toxicity tests were performed to contrast responses of *Daphnia magiza* Strauss, *Hyalella azteca* Saussure, *Chironoinus tentaiis* Fabricius, and *Pimephales promelas* Rafinesque to chelated-copper herbicides: Clearigate®, Cutrine® - Plus, and copper sulfate. Organisms were exposed in nonrenewable static systems to determine short-term toxicity (48-h LC_{50}s) and bioavailability (after 7 d) of these copper compounds. For both Clearigate® and Cutrine® - Plus, the microcrustacean *D. magna* (2.55 and 8.61% mortality/µg Cu/L, respectively) was ≥12 and ≥ 25 times more sensitive than *H. azteca*, *C. tentans*, and *P. promelas*. *H. azteca* and *P. promelas* were approximately 5 times more sensitive to Cutrine® - Plus *(0.35* and 0.275% mortality/µg Cu/L, respectively) than *C. tentans* (0.062% mortality/µg Cu/L). *D. magna* was also the most sensitive test organism to copper sulfate (5.07% mortality/µg Cu/L). As compared to *D. magna*, differences in copper sulfate potency varied by a factor of 2-90(2.85% mortality/µg Cu/L for *P. promelas* to 0.057% mortality/µg Cu/L for *C. tentans)*. The concentrations of bioavailable free ion Cu^{2+} and associated complexes decrease during a one week period based on responses of all four test organisms. Exposure-response slopes, in conjunction with point estimates, are useful for extrapolations and for predicting effects of herbicides to aquatic populations.

PWA018 Bioconcentration of Pyribenzoxim in Killifish (*Oryzias latipes*). Kim,J.H.* College of Agriculture & Life Science, Seoul National University, Suwon, Korea, Yeom,D.H, Kim,K., Kim,J.C., Lee,S.K. Kim,Y.H. Korea Research Institute of Chemical Technology, Taejon, Korea, Pyribenzoxim is a new herbicide developed by LG Chemical

Ltd, Korea, acting as a ALS(acetolactase synthase) inhibitor. Prior to bioconcentration study the acute toxicity of pyribenzoxim to killifish was determined (>100 mg/l)in a 96h semi-static test. Bioconcentration study utilized a flow-through exposure system in which the test organisms were continuously exposed to two sublethal concentrations of pyribenzoxim (84 and 789 ppb) for 20 days, followed by a depuration period for 5 days in clean water. Periodic sampling of water and whole fish were made throughout the uptake (days 0, 1, 3, 7, 10, 14, 18, 20)and depuration phase (days 21, 22, 25). In fish, the concentration of pyribenzoxim reached at steady-state from days 7 to days 20, maintaining at 7.0 ppm for low concentration test (84ppb) and at 45.7ppm for high concentration test (789ppb). BCF of pyribenzoxim was determined to be 70 by averaging BCFs of five points(days 7, 10, 14, 18, 20) on steady-state from each concentration.

PWA019 Hepatopathology of the drinking water disinfection by-product, dichloroacetic acid, in the Japanese medaka fish model. Lopez, L., Toxicology Dept., and Law, J. M.., College of Veterinary Medicine, North Carolina State University, Butterworth, B.E., Chemical Industry Institute of Toxicology. Dichloroacetic acid (DCA), a drinking water disinfection by-product is carcinogenic to rats and mice. Appropriate risk assessments are limited because the mechanism of DCA induced cancer is unknown. The Japanese medaka (*Oryzius latipes*) small fish model, is used increasingly for carcinogenicity testing due to its ease of maintenance, and carcinogen sensitivity. In this initiation-promotion study, 6-week old medaka were or were not exposed to diethyl-nitrosamine (DEN), followed by chronic DCA exposure (5.0, 50.0, 100.0 or 500.0 mg/l) in ambient water for 26 weeks. Results thus far, strengthen the medaka's role as a suitable species in carcinogenicity testing. Cell proliferation was quantified using bromodeoxyuridine (BrdU) exposure (50 ppm) in the ambient water three days prior to sacrifice to label cells in S-phase. As early as week three, concentration dependent diffuse hepatocellular changes included cytomegaly, marked cytoplasmic vacuolation, karyomegaly, nuclear atypia, scattered areas of hepatocellular necrosis, cell loss, increased cell proliferation and excessive intracellular glycogen deposition. Total cellular changes over 13 weeks are similar to those seen in rodents exposed to DCA. An elevated labeling index (LI) was also characterized by an increase in animal to animal variability with strong and weak responders. DEN treatment increased LI at all time points while DCA slightly reduced the DEN induced LI. Not until week 13 did a dramatic increase in LI occur in the DEN/DCA 50 mg/l and DCA 100 mg/l groups. Histopathology, labelling index, and apoptotic rate are complementary endpoints in understanding underlying tumor formation mechanisms in the medaka model.

PWA020 Toxicological Assessment of Chloroform Using Medaka Fish. Toussaint, M.W.*, GEO-CENTERS, INC., Fort Detrick, MD; Gardner, H.S., USACEHR, Fort Detrick, MD; Beaman, J.R., GEO-CENTERS, INC., Fort Detrick, MD; Rosencrance, A.B., USACEHR, Fort Detrick, MD; Wolfe, M.J., Experimental Pathology Laboratories, Inc., Herndon, VA. The effects of the common drinking water disinfection byproduct chloroform on fish have been assessed using Japanese medaka fish (*Oryzias latipes*). Assessments were performed during the course of the nine-month continual aqueous dosing of medaka with chloroform (0.015, 0.15, or 1.5 mg/L nominally). There were four replicate aquaria per treatment. Test solutions were replenished at a rate of 100 mL/min in the 5 gallon dosing aquaria. Actual concentrations of chloroform in tank samples were determined weekly. After six months of exposure, an interim histopathology sacrifice was performed, and immune system function was assessed. At nine months, liver tissue concentration of chloroform was determined. The remainder of the fish on test were sacrificed at nine months for histopathologic evaluation. Measured analysis of the three chloroform treatments revealed mean measured concentrations to be: 0.018 ±0.004, 0.151± 0.034 and 1.46 ±0.320 mg/L. Immune system function was determined through hematological analysis. Hematocrit and leukocrit, as well as pronephric cell counts and cell viability, of treated fish were not different from controls. Chloroform tissue concentrations ranged from approximately 2 to 14 times less than the tank concentrations. At 0.151 mg/L actual tank concentration, fish tissue concentration averaged 0.083±0.071 mg chloroform per mg fish liver. At 1.46 mg/L actual tank concentration, fish tissue concentration averaged 0.107±0.077 mg chloroform per mg fish liver. No chloroform was detected in the hepatic tissue from the 0.018 mg/L external concentration tanks. Mortality was <4% in all groups, and there were no treatment-related significant differences found in fish length or weight. Although treatment-related liver carcinogenicity was not seen at six or nine months, gall bladder lesions and bile duct abnormalities were seen in the fish treated with 1.46 mg/L chloroform.

PWA021 Comparison of the Aquatic Toxicity of 2-(Thiocyanomethylthio) Benzothiazole and Selected Degradation Products with *Daphnia magna* and Microtox™. Drake, K. D.*, Zhou, X., Buckman Laboratories International, Inc., Memphis, TN; Blanner, P., Maier, K. J., University of Memphis, Memphis, TN. Laboratory tests were conducted to compare the aquatic toxicity of 2-(thiocyanomethylthio) benzothiazole [TCMTB] and several of its benzothiazole-based degradation products. TCMTB is a fungicide used in a number of industrial processes. Aquatic toxicity tests have shown that TCMTB is highly toxic to aquatic organisms. Previous publications have reported the potential for environmental exposures and the routes of degradation. The following compounds were evaluated: TCTMB, 2-mercaptobenzothiazole, hydroxybenzothiazole, methylthiobenzothiazole, 2-benzothiazole sulfonic acid. The *Daphnia magna* tests were performed under static renewal conditions with measured concentrations. The Microtox assay was conducted per the manufacturer's recommendations. The results of the invertebrate testing confirm TCMTB is highly toxic to aquatic organisms. The benzothiazole-based degradation products are significantly less toxic than the parent compound. The Microtox results are similar to the *Daphnia magna* testing, these show TCMTB as being the most toxic with the benzothiazole-based degradation products being significantly less toxic.

PWA022 Assessment of toxicity of crude and weathered oil in rockfish, *Sebastes schlegeli* and Microtox. Kim, J-.S.*, Hwang, I.Y., Bae, C.H., Yi. M.A., Kim, Y.E., Inje University; Park, K.H., Kunsan National University, Korea. Marine pollution by oil and petroleum hydrocarbons have received much attention due to mainly their lethal and sublethal effects on marine organisms. In this study, Arabian medium and Kaphchi crude oil were artificially weathered for 2 weeks in our special designed oil-weathering chamber to assess the change of its toxic effects on rockfish, *Sebastes schlegeli* and Microtox. Rockfish was exposed to water soluble fraction(WSF) and dispersed oil fraction(DOF) of crude and weathered oil for one week. As biomarkers of exposure, hepatic phase I (ethoxyresorufin-O-deethylase,EROD; arylhydrocarbon hydroxylase, AHH) and phase II (glutathione –S-transferase, GST) enzyme activities were meseasured. In addition, change of Microtox EC50 of oil layer was monitored during weathering process. EROD and AHH acitivities of the fish exposed to WAF(100%) were induced 4.9-, and 3.4-folds, respectively, while to DOF(300ppm), 16.8-, and 11.0-folds, respectively. GST activity was significantly reduced by exposure to DOF. However, GST was not modulated by exposure to WAF. EC50 of Microtox for oil layer was drastically changed from 20ppm to 3000ppm(Arabian medium crude oil) and 7500ppm(Kaphchi crude oil) for 2 weeks in the presence of metallic copper. These findings indicates that the modulation of phase I and II enzyme activities and Microtox EC50 were good indicators to evaluate the impact of the oil contamination on some marine organisms.

PWA023 Are Aromatic Hydrocarbons the Primary Determinant of Petroleum Toxicity to Aquatic Organisms? Barron, M.G.,* Podrabsky, T., Hagler Bailly Services, Boulder, CO; Ogle, S., Pacific Eco-Risk Labs, Martinez, CA; Ricker, R.W., California Department of Fish and Game/Oil Spill Prevention and Response, Sacramento, CA. Aromatic components of oil, particularly polycyclic aromatic hydrocarbons (PAHs), are generally assumed to be the toxic fraction of spilled petroleum. We evaluated this assumption by analyzing the chemistry and toxicity of water accommodated fractions (WAFs) prepared from three environmentally weathered middle distillate oils (C10 to C32 hydrocarbon range) differing in aromatic content. Oil toxicity was determined in short-term growth and survival tests with the mysid shrimp *Mysidopsis bahia*. Median lethal concentrations (LC50s) ranged from 0.9 to 1.5 mg/L total petroleum hydrocarbons (TPH), and growth-inhibiting concentrations (EC20s) ranged from 0.13 to 1.1 mg/L TPH. Toxicity of the three oils expressed as $\mu g \sum PAH/L$ (sum of 41 PAH analytes; parent and alkyl homologs) ranged from 2.2 to 12 $\mu g/L$ (0.02 to 0.07 μM) (LC50s) and 0.32 to 5.7 $\mu g/L$ (0.002 to 0.04 μM) (EC20s). The test oil with the lowest aromatic content had the greatest toxicity when expressed as mg/L TPH, $\mu g/L \sum PAH$, or $\mu g/L$

∑naphthalenes in WAF. The test oil WAF with the highest toxicity also had the lowest concentrations of single PAH analytes measured at concentrations greater than the detection limit. The results of this study demonstrated that low aromatic content oil can be highly toxic and that PAHs were not the major determinant of the toxicity of the three weathered middle distillate oils.

PWA024 Influence Of Dispersants On Bioavailability And Trophic Transfer Of Phenanthrene To Algae And Rotifers. Wolfe, M.F.*, Schwartz, G.J.B., Singaram, S., Mielbrecht, E.E., Tjeerdema, R.S., University of California, Santa Cruz, California; Sowby, M.L., California Department of Fish and Game, Sacramento, California. The objective of this research was to determine the uptake, bioaccumulation, and depuration of [14C]phenanthrene from Prudhoe Bay crude oil (PBCO) dispersed with Corexit 9527 (DO) compared with undispersed preparations of the water-accommodated fraction (WAF) of PBCO by primary levels of a representative marine food chain. Isochrysis galbana, a primary producer, and Brachionus plicatilis, a primary consumer were used to compare direct aqueous (AQ) exposure with combined aqueous and dietary (AQ&D) exposure. Results showed uptake of phenanthrene by algae was increased significantly ($p<0.05$) from 102 to 172 nmol eq phenanthrene/g algae in the presence of dispersant. No significant difference ($p>0.05$) in uptake of phenanthrene was observed in rotifers in the presence of dispersant, however, depuration of phenanthrene significantly ($p<0.01$) decreased following DO exposures. Uptake of phenanthrene increased significantly ($p<0.05$) via trophic transfer in WAF, from 51 to 87 nmol eq phenanthrene/g rotifers in AQ and AQ&D, respectively, and similarly, from 57 to 130 nmol eq phenanthrene/g rotifers, in DO exposures. Retention of tissue residues of phenanthrene during depuration were further enhanced with trophic transfer. Depuration rate constants confirmed observed differences coincident with different routes of exposure. Bioaccumulation of [14C]phenanthrene decreased significantly in the presence of dispersant in both algae and rotifers while increasing significantly with trophic transfer in both WAF and DO exposures. This research demonstrated dispersants altered uptake, bioaccumulation, and depuration processes of phenanthrene in representative primary species which may in turn modify bioavailability and bioaccumulation at higher trophic levels.

PWA025 Aquatic Toxicity of Three Major Gasoline Blending Streams in Surface Waters. Rausina, G.[1]*, BenKinney M.[2], Breglia, R.[3], Burnett, D.[4], Dmytrasz, B[5], Koschier, F.[6], Lapan, C.[6], Leak, T.[7], Podhasky, P.[8], Schreiner, C.[2], and White, R.[1]; [1] Chevron; [2] Mobil; [3] BP; [4] AMOCO; [5] Texaco; [6] ARCO; [7] ABC Labs; [8] Petroleum Product Stewardship Council. The inherent aquatic toxicity of three major gasoline blending streams (i.e., light alkylate naphtha, light catalytically cracked naphtha and light catalytically reformed naphtha) was assessed in sealed test vessels to maintain stable test levels. Test organisms were exposed to individual water accommodated fractions (WAFs) of each gasoline blending stream using procedures set forth in ASTM D6081 draft methods, except the WAF generation vessels were tightly covered, with minimal headspace to reduce volatilization of hydrocarbons. Freshwater and marine fish (fathead minnow, *Pimephales promelas* and silverside minnow, *Menidia beryllina*) and invertebrates (cladoceran, *Daphnia magna* and mysid shrimp, *Mysidopsis bahia*) were tested under static-renewal conditions with daily replenishment of freshly prepared WAFs. Freshwater green alga (*Selenastrum capricornutum*) tests were static non-renewal. Measured test concentrations based on total levels of representative analytes in each gasoline blending stream were determined by purge-and–trap gas chromatography/FID. Inherent toxicity values form the basis for initial hazard assessments of possible exposure scenarios as well as for product registration and labeling. However, they can over-predict gasoline blending stream toxicity in any ecological hazard assessment where factors that govern environmental fate (i.e., volatility, dissolution kinetics and biodegradability) of gasoline hydrocarbons have not been accounted for. Hence, in many exposure scenarios there are adequate margins of safety between transient exposures of gasoline hydrocarbons in surface waters and the aquatic toxicity thresholds of the three major blending streams.

PWA026 Toxicity of Methyl Tertiary-Butyl Ether (MTBE) to Selected Marine and Freshwater Organisms. Drottar, K.R.*, Mank, M.A., Palmer, S.J., Sulaiman, M.W., Springer, T.S., and Krueger, H.O., Wildlife International, Ltd., Easton, MD; Steen, A.E., American Petroleum Institute, Washington, D.C.; Rausina, G.A., Chevron Research & Technology Co., Richmond, CA; Wong, D.C.L., Shell Development Company, Houston, TX; Arnold, R.W., Exxon Biomedical Sciences, Inc., East Millstone, NJ. MTBE is a fuel oxygenate used extensively to enhance the octane rating of fuels and to reduce tailpipe emissions. Acute and chronic toxicity tests of MTBE were performed on a number of marine and freshwater organisms to obtain toxicity data required for calculation of ambient water quality criteria. These studies are part of a collaborative effort involving a variety of industry representatives, the EPA, and testing laboratories. Acute toxicity tests were performed with sheepshead minnow (*Cyprinodon variegatus*), three-spine Stickleback (*Gasterosteus aculeatus*), blue crab (*Callinectes sapidus*), *Daphnia magna*, Mysid shrimp (*Mysidopsis bahia*), grass shrimp (*Palaemonetes pugio*), oyster (*Crassostrea virginica*) and the marine alga *Skeletonema costatum*. Chronic tests were performed on Mysid shrimp and *Daphnia magna*. All flow-through tests were performed in a diluter system where MTBE and dilution water were thoroughly mixed in a closed-headspace mixing chamber before flowing into test chambers. MTBE concentrations were analytically determined in all tests. Responses of animals in chronic studies were evaluated by calculating NOEC and IC25 values. The values of the various endpoints are compared to MTBE concentrations that have been measured in urban storm-water runoff and surface waters.

PWA027 Impact of Trifluoroacetic Acid (TFA) on the Fathead Minnow (*Pimephales promelas*) in Outdoor Microcosms Hanson, M.L.*, Solomon, K.R., Centre for Toxicology, University of Guelph, Canada; Muir, D.C.G., National Water Research Institute, Burlington, Canada; Mabury, S.A., Dept. of Chemistry, University of Toronto, Canada. Trifluoroacetic acid (TFA) is the major degradation product of several hydrofluorocarbons and hydrochlorofluorocarbons being used to replace the ozone-depleting chlorofluorocarbon refrigerants. This compound is highly persistent and is projected to reach concentrations of up to 100μg/L in the next 30 years in some regions of the world. During the summer of 1997, 12m^3 microcosms located at the Guelph Microcosm Facility were dosed with TFA at concentrations of 10, 100, 300 and 1000μg/L. Prior to dosing, the microcosms were stocked with 8 mating pairs of fathead minnows (*Pimephales promelas* Rafinesque). Mortality and egg-laying were monitored daily over the exposure period. The study was completed after 12 weeks with the removal of surviving adults and juveniles. Overall, TFA reduced mortality in adult fatheads compared to controls. This may be due to an antimicrobial or antifungal action. Adult fish at 1000μg/L TFA showed a significant weight loss after 12 weeks of exposure. Juvenile survival was impaired, with a significant decline in survivorship at concentrations of 100 and 1000μg/L TFA. Total biomass of the surviving fathead juveniles appeared to be unrelated to TFA concentration and is likely a result of competition stress. This study shows TFA has a potentially detrimental role in aquatic ecosystems. Ongoing laboratory experiments related to this study involving aconitase and citric acid levels in fathead minnows will be discussed.

PWA028 Toxicity Evaluation of Two Fuel Oils Upon Five Different Marine Species. Reynier, M.V., Marroquim, A.C., Kraus, L.A.S., Universidade Federal do Rio de Janeiro, Rio de Janeiro, RJ, Brazil; Maurat, M.C.S., Universidade Federal Fluminense, Niterói, RJ, Brazil; Gabardo, I., Maia, D.B.M., Vital, N.A.A., Centro de Pesquisas da Petrobras - Diquim/Sebio, Rio de Janeiro, RJ, Brazil. The toxicity of two crude oils was evaluated upon four marine species and the Microtox® Acute Test System. The algae *Champia parvula*, the sea urchin *Lytechinus variegatus*, the marine luminescent bacteria *Vibrio fischeri*, the mysid *Mysidium gracile* and the fish *Poecilia vivipara* were exposed to the water-accomodated fractions (WAFs), generated from the mixture of individual loading rates. The required amount of oil was weighted using a syringe and added to the water surface. The solution was mixed for 60 seconds at 13.000 rpm. The oils were fairly viscous, determined as semi-solids and their characterizations were done by GC-FID, TLC-FID and UV-fluorescence methods. The total hydrocarbon content in the test solutions was measured using an oil content analyzer (Horiba OCMA-220 model). The results of the two week static renewal algae growth test and the fertilization and embryological development tests with the sea urchin (NOEC, OEC) were compared. The survival of the mysids, fish (static 96 hours nonrenewal assays - LC50) and bioluminescence inhibition of marine bacteria (15 minutes - EC50) was also compared. The preliminary results indicated that the sensitivity of *C. parvula* and *L. variegatus* (embriological tests) were the same and the rank from least to most sensitive in the acute tests were: MICROTOX < *P. vivipara* < *M. gracile*.

PWA029 Field Evaluation of Ecotoxic Impacts to Clear Creek, Houston, Texas. Plaza, A.*, Jacob, T., Nwankwo, C. and Howard, C.L., University of Houston – Clear Lake, Houston, TX USA. In the Houston – Harris County region of Texas, most natural streams have been channelized over the past 50 years in an attempt to control flooding. Clear Creek, a 30-mile long meander emptying into Clear Lake and Galveston Bay, is the notable exception. Although still technically in its natural state, Clear Creek has been subjected to a number of toxic inputs from sources that include the Brio Superfund Site and runoff from pastureland and residential areas. In the study described here, we collected water, sediment and biota from two reference sites in Armand Bayou and 12 sites along the 14-mile reach of Clear Creek under consideration for channelization. Water, sediment and selected fish were analyzed for heavy metals (by ICP and atomic absorption spectroscopy) and PAHs (by GCMS). Stream water toxicity was determined using chronic exposures to *Daphnia pulex* and Microtox, and benthic macroinvertebrate diversity was evaluated. Our results indicate that sediments are contaminated in specific places along Clear Creek and toxic impacts to test species, as well as the natural community, are measurable.

PWA030 Simple and Rapid Extraction of Polycyclic Aromatic Hydrocarbons Collected on Polyurethane Foam Adsorbent. Maddalena, R.L.*; N.Y. Kado, University of California, Davis, CA 95616; T.E. McKone, Lawrence Berkeley National Laboratory and University of California, Berkeley, CA 94720. A single pass flow through extraction (FTE) method for polycyclic aromatic hydrocarbons (PAHs) that have been collected on poly-ether type polyurethane foam (PUF) is described and demonstrated. The method is based on the principles of trace enrichment and solid-phase extraction where the analyte of interest is collected or concentrated on the expanded PUF matrix. Following collection, the PUF is compressed to reduce the solvent requirement and a single pass gravity fed mobile phase extraction is used to remove the analyte. Two extraction mobile phases were tested (dichloromethane, DCM and a 10% (V:V) mixture of acetone in hexane, AH). Quantitative recoveries of a set of 18 PAHs were achieved with less than 4 ml of DCM or 6 ml of AH. Average standard recoveries of 18 spiked PAHs were 97% and 104 % for the DCM and the AH extractions, respectively. The semi-volatile fraction of PAHs in an environmental sample of a complex mixture (diesel exhaust) were also quantitatively extracted using the FTE method. Finally, the FTE method was used to extract samples collected from an exposure chamber that was designed to measure the transfer of semi-volatile atmospheric pollutants into vegetation. Total variance reported as percent relative standard deviation in the measured steady-state atmospheric concentrations of pyrene, fluoranthene, anthracene and phenanthrene were 3.4%, 10.5%, 8.5% and 4.4%, respectively, thus demonstrating the excellent precision of the method. The FTE method is simple, fast and effective for extracting PAH that has been collected on PUF adsorbent.

PWA031 Extraction of Aqueous Phase Polycyclic Aromatic Hydrocarbons (PAHs) into Microliter Volumes of Solvent. McEachern, P. R.* and Foster, G. D., George Mason University, Fairfax, Virginia. The liquid-liquid extraction (LLE) of hydrophobic organic contaminants in water is a highly efficient preconcentration step in trace analysis. Most low detection limit LLE methods (i.e., < 1 ng/L) use large quantities (> 200 mL) of organic solvent to extract organic contaminants from 50 L or more of water for subsequent analysis. The process requires considerable solvent volume reduction and handling which leads to considerable analyte losses in some cases as well as large volumes of solvent waste. A new liquid-liquid microextractor has been developed which uses less than 10 microliters of solvent to extract up to 500-mL of water, providing detection limits similar to large volume LLE. The micorextractor consists of a modified 25 microliter syringe fitted to a 20-mL customized flow-through mixing chamber. Dissolved phase PAH have been extracted from water into a solvent droplet formed at the tip of the syringe needle. An aliquot of the solvent was injected directly into a gas chromatograph for analyte quantitation without the need to concentrate the solvent. A microextractor has been constructed and evaluated with the objectives of (1) reducing both the sample and solvent volume used per extraction, and (2) optimizing solvent physical properties, sample flow rate, and mixing rate. Preliminary results involving the performance of the microextractor have shown that the device follows LLE theory in most cases and has been found to be very efficient in the aqueous phase extraction of the most hydrophobic PAH.

PWA032 An Overview of the Results of Several Comparisons of Lipid-Containing Semipermeable Membrane Devices (SPMDs) and Biomonitoring Organisms for Assessing Organic Chemical Exposure. Huckins, J.N.*[1]; Prest, H.F.[2]; Petty, J.D.[1]; Roe, T.I.[3]; Meadows, J.C.[1]; Echols, K.R.[1]; Lebo, J.A.[1]; Clark, R.C.[1]. USGS, Columbia, MO[1]; Long Marine Laboratory, Santa Cruz, CA[2]; and Norsk Hydro, Bergen Norway[3]. In several monitoring programs, SPMDs are being proposed as an addition to or replacement for biomonitoring organisms. To facilitate this transition, an improved understanding is required of the relative performance of SPMDs and organisms with regard to their accumulation of organic contaminants. In this work we examine the results of three side-by-side SPMD-biomonitor exposures and explore similarities and differences in chemical exchange kinetics and thermodynamics. Organisms used in these studies include oysters (*Crassostrea gigas*), mussels (*Mytilus edulis*), and fish (*Salmo trutta*). Target compounds were polycyclic aromatic hydrocarbons (PAHs) and polychlorinated biphenyls (PCBs); in all exposures SPMDs concentrated as many or more chemicals than the biomonitors. In terms of uptake kinetics or the relative uptake rates of the four matrices the following sequence appears to apply: oysters > SPMDs > fish > mussels. However, ranking is complicated by the choice of the method used to present the data, i.e., wet, dry, or lipid weighted. Detailed results of these studies plus other relevant research results will be discussed.

PWA033 Use of PAH Diagnostic Ratios for Source Identification: A Critical Review. Graham, S.E.*, Shea, D., North Carolina State University, Raleigh, NC. Various ratios of polycyclic aromatic hydrocarbons (PAHs), such as the fossil fuel pollution index, have been used over the past two decades to help identify sources of PAHs to the environment. We critically reviewed the use of PAH distribution ratios as a diagnostic tool for identifying sources of PAH to the aquatic environment. In compiling a large database of diagnostic ratios, we reviewed both primary sources (crude oils, petroleum products, specific soots, etc.) and secondary sources (sewage effluent, street dusts, rivers, etc.). We also reviewed PAH ratios reported for receptors, primarily air particulate and sediment, from both urban and remote locations. Diagnostic ratios included, parent compound distributions (PCDs), parent compound isomer ratios (PCIR), alkylated-to-parent compound ratios (e.g. methyl-phenanthrane/phenanthrene or MP/P), among others. Individual source discrimination was accomplished using principal component analysis (PCA) for fresh petroleum, weathered petroleum and combustion type PCDs. Significant positive correlations ($p<0.05$) were revealed among the majority of the PCIRs, with pyrogenic sources generally containing higher values. This suggests that increased PCIRs determined at a given receptor would indicate increased combustion derived PAH. We also found that MP/P was negatively correlated ($p<0.05$) to the PCIRs, supporting the observation that petrogenic residues contain a higher degree of alkylation. Our analysis provides a broad base of support for the use of PAH diagnostic ratios, but also reveals clear limitations to their use. The main limitation is the inability to identify primary PAH sources in systems containing complex mixtures from multiple sources. However, it is possible to identify secondary sources in many complex systems, providing regulators and resource managers with a powerful tool to identify and apportion sources of PAH to the aquatic environment.

PWA034 Evaluation of a Fluorometric Screening Method for Estimating Total PAH Concentrations in Sediments. Peterson, G.S.*, Natural Resources Research Institute, Duluth, MN; Axler, R.P., Natural Resources Research Institute, Duluth, MN; Lodge, K.B., University of MN, Duluth, MN; Crane, J.L., Minnesota Pollution Control Agency, St. Paul, MN. A fluorometric screening method was used to estimate total polycyclic aromatic hydrocarbon (PAH) concentrations in sediments collected from the St. Louis River Area of Concern (AOC) in northeastern Minnesota. Sediments were collected as part of a Regional Environmental Monitoring and Assessment Program (R-EMAP) study to assess sediment quality in the AOC. The screening method was calibrated using a PAH surrogate standard consisting of eight PAHs commonly found in the St. Louis River system, at their approximate proportions. Estimated PAH concentrations were compared to GC/MS measured "true"total PAH concentrations to evaluate the overall predictive success of the screening method. Regression analysis of estimated versus "true" PAH concentration yielded an r^2 of 0.70 (n=56) and 0.73 (n=33) for samples collected in 1995 and 1996 respectively. The data sets were analyzed separately because the intercepts of the two regression analyses were determined to be significantly different. Screening estimates were also compared to the GC/MS measured values relative to four different sediment effect concentrations (SECs) for total PAHs. This comparison was done to determine the rates of false positive (type I error) and false negative (type II error) predictions associated with the screening method. The rate of false positive predictions was shown to decrease as the SEC criteria value increased, while false negative rates remained consistently low, below 7%. Methodological recommendations which led to the improved prediction of both high and low PAH samples will be presented.

PWA035 Determination of Phthalate Esters in Environmental Samples by Mass Spectrometry Based Analytical Methods. Lin, Z. P.*[1,2], Michael G. Ikonomou[2] and Frank Gobas[1], [1]School of Resource and Environmental Management, Faculty of Applied Sciences, Simon Fraser University, Burnaby, BC, Canada; [2]Ocean Chemistry, Institute for Ocean Sciences, Sidney, BC, Canada. Most analytical methods associated with the analysis of phthalates use GC-ECD or GC/LRMS detection. Phthalates fragment readily under GC/LRMS conditions and as a result the molecular ions are not present in the mass spectra. In most applications the ions monitored for quantitative work are fragment ions common to most phthalates such as m/z 163 and 149. The degree of fragmentation depends on the type of MS instrumentation and ionization conditions used. In this work we have examined a number of mass spectrometric techniques for the quantitative determination of phthalates in environmental samples. Test solutions containing 18 of the most common phthalates were analyzed by GC/LRMS using +ve EI, +ve CI, and -ve CI, and also by GC/HRMS (+ve EI), LC/ESI-MS (both +ve and -ve ionization modes) and GC-ECD. The objective was to examine these techniques using modern instrumentation in terms of specificity, detection limits and quantitation precision using low and high concentration test solutions. The advantages and limitations of each technique will be discussed and the data will be compared against corresponding literature reported values. The detection limits of the overall analytical method using the preferred detection technique was assessed using spiked environmental matrices (water, tissue, sediment and sludge).

PWA036 Separation of a technical mixture of p-nonylphenol into component isomers by HPLC. Gundersen, J.L.* U.S. EPA, Atlantic Ecology Division, Narragansett, RI, USA. p-Nonylphenol (NP) is the ultimate degradation product of nonylphenol polyethoxylate surfactants and has been reported to be an endocrine disrupter. It is composed of numerous structural isomers resulting from the various branching patterns of the C_9 group. High resolution GC-MS has identified 22 isomers in a technical mix of NP. In most HPLC analyses, nonylphenol elutes as a single, broad peak. In the method described here, HPLC using a graphite carbon column resulted in the resolution of NP into 10 peaks or groups of isomers. Separation was achieved by elution with 1% acetic acid in water and acetonitrile. This method can be used to fractionate NP based on structure and assess the potential for different isomers (or groups of structurally similar isomers) to act as endocrine disrupters.

PWA039 ^{19}F MAS NMR spectroscopic studies on the degradation of fluorinated xenobiotics in soil matrices: A new approach to the study of pollutant biotransformations in intact samples. Green, N.A.*, Lindon, J.C. and Nicholson J.K.. Imperial College of Science Technology and Medicine, London, UK. ^{19}F MAS NMR spectroscopy was used to monitor the qualitative and quantitative transformations of a series of trifluoromethyl anilines and phenols in whole soil samples. Detection limits of <100mg kg^{-1} soil for the technique was established, demonstrating the ability to carry out investigations at environmentally relevant levels. The degree of dispersion of the compounds within the soil were determined by their distributions throughout the sample, giving an indication of their homogeneity. The ^{19}F line-width was found to significantly broaden over time indicating that the compounds were being incorporated into the soil organic matter and therefore molecular motion was becoming increasingly restricted. The restricted molecular motion causing such characteristic broadening of the peaks would be due to chemical anisotropy effects and dipolar couplings. Interactions of the compounds and the soil organic matter were investigated to determine rates of adsorption and desorption. A comparison between the ^{19}F MAS NMR spectroscopic data and established approaches to determine adsorption isotherms was carried out. From these preliminary studies it can be seen that ^{19}F MAS NMR spectroscopy will become an important technique for investigating fluorinated xenobiotic transformations in soil.

PWA040 Use of Semipermeable Membrane Devices For Monitoring Non-Persistent Pesticide Concentrations in South Florida Canals. Strozier, E.D., NOAA/NOS, Charleston, SC.; Kucklick, J.R., NIST Charleston Lab, Charleston, SC; Scott, G.I., NOAA/NOS, Charleston, SC. Traditionally grab water samples have been used to monitor contaminant levels in water systems. However grab sampling methods have several disadvantages. First, grab water samples only represent a single point in time. Grab sampling also requires the collection of large sample volumes and the extraction, preparation of these samples is labor intensive and time consuming. Recently the use of Semipermeable Membrane Devices (SPMDs) has been proposed as an alternative or addition to grab sampling methods. SPMDs sample the water continuously until they reach equilibrium. During the period from February 1997 to February 1998, we have conducted three separate field trials with SPMDs to assess their usefulness in replacing grab sampling for assessing water concentrations of non-persistent pesticides. For each trial, SPMDs were deployed for five days at three to five sites along the C-III canal in South Florida. Daily water samples were also collected at each site during the study periods. At two of the sites additional composite water samples were taken by an automatic sampler at 6h intervals. Filtered water was extracted using ENV+ solid phase extraction cartridges, SPMDs were extracted by dialysis in hexane. All extracts were analyzed by capillary GC with ECD and/or NPD. Total Endosulfan levels between 1.9 and 130 ng/L were measured in grab water samples. Water contaminant levels determined from SPMDs were between 7.3 and 192 ng/L. These results indicate that SPMDs may be a viable substitute for grab sampling.

PWA041 Solid-Phase Microextraction of Metribuzin, Chlorothalonil, Endosulfan and Fenvalerate in Runoff Water from Tomato Production. McConnell, L.L.*, USDA, Agricultural Research Service, Beltsville, MD; Nochetto, C.B., University of Maryland, Chesapeake Biological Laboratory; Rice, P.J., USDA, Agricultural Research Service, Beltsville, MD. A solvent-free, fast technique to quantitate pesticide concentrations in agricultural run-off water was developed for a field plot experiment to determine pesticide loss from two different tomato production systems. Low volume water samples (1.5 ml) were extracted with agitation and analyzed after filtration to 0.7 µm using automated solid-phase microextraction (SPME) coupled with capillary gas chromatography and electron-capture detection. Studies in the lab showed a linear response of the different pesticides over a wide concentration range (ppt-ppb). The standard curves had an R value of 0.997 or higher for all the pesticides analyzed (metribuzin, chlorotalonil, α-endosulfan, β-endosulfan, and esfenvalerate). Two different fibers were used: 85 µm polyacrylate for metribuzin and 100 µm polydimethylsiloxane (PDMX) for the remaining compounds. The method was also evaluated with respect to the limit of detection (LOD), method detection limit (MDL) and precision (RSD). Surrogates (PCB 204 and PCNB) were added to the water sample before each extraction to check recoveries of the method and status of the fibre. The method represents a feasible alternative to normal solid-phase extraction techniques for projects where concentration levels are > 12 ppb for metribuzin, chlorothalonil, and esfenvalerate and > 30 ppt for endosulfan, and where large numbers of samples are processed.

PWA042 A Simple Procedure for the Analysis of Pesticides in Aqueous and Biotic Matrices. J.B. Belden* and M.J. Lydy, Department of Biological Sciences, Wichita State University, Wichita Ks. Determining the presence and concentrations of pesticides in the environment is difficult and expensive due to the large variety of chemicals applied. Using standard EPA protocols, several extraction and analytical techniques, requiring a variety of expensive equipment and chlorinated solvents, would be necessary to monitor water samples for pesticides which are in different chemical families. Methods are not available for many contemporary pesticides, while still fewer methods are available for extraction and cleanup of biota. An analytical method involving a single extraction and analytical technique was developed for the determination of selected chlorinated insecticides, organophosphate insecticides, organophosphate metabolites, carbamate insecticides, and triazine herbicides. Aqueous samples were extracted utilizing solid phase extraction techniques. Biota samples were extracted by sonication with organic solvent and the resulting extracts were "cleaned" by normal phase column chromatography with florisil as the stationary phase. The resulting extract was concentrated and analyzed by a gas chromatograph coupled with a nitrogen-phosphate detector. This method provides a low cost, environmentally friendly analysis which is sensitive enough to determine the presence of pesticides within complex matrices at ecologically relevant levels.

PWA043 Comparison of H4IIE TCDD-TEQs in River Sediments following Sample Preparation by Accelerated Solvent Extraction (ASE) or Soxhlet techniques. McFarland,V.A., USAE Waterways Experiment Station, Vicksburg, MS; McCant, D.D.*, ASci Corporation, Vicksburg, MS; Inouye, L.S., ASci Corporation, Vicksburg, MS. Extractions were carried out on six 10 g aliquots of Saginaw River sediments using either Accelerated Solvent Extraction (ASE) or Soxhlet techniques. The extracts were then assayed with cultured Reuber H4IIE rat hepatoma cells standardized against 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) measuring ethoxyresorufin O-deethylase [EROD] activity quantitated using a fluorescence microplate reader. Results for ASE and Soxhlet extraction techniques were 215 ± 77 and 202 ± 84 pg TCDD-EQ/g sediment, respectively. ASE extractions required only 30 minutes as opposed to 18 to 24 hours per sample for Soxhlet extractions. Additionally, ASE required far smaller volumes of solvent (50 mls as opposed to 250 mls with Soxhlet). These advantages coupled with one step cleanup furthur reduced turn around time by 24 hours for ASE extractions. Reported TCDD ^{13}C surrogate recoveries for ASE and Soxhlet were 72 and 68 %, respectively. Comparable results obtained with far more rapid throughput and reduced solvent requirements represent considerable advantages of ASE over Soxhlet extraction. These are counterbalanced by the high cost of initial investment in ASE equipment.

PWA044 Application of MS/MS Techniques in Trace Analyses of Complex Environmental Samples. Morrall, S.W.*, Begley, W.M., and Simonich, S.L., The Procter & Gamble Company, Cincinnati, OH; Robaugh, D.A., Midwest Research Institute, Kansas City, MO. Trace analyses in complex environmental matrices has often relied on high resolution capillary GC to isolate analytes from an obscuring chemical background, prior to identification and detection by mass spectrometry (MS). Electrospray ionization (ESI) has enabled the routine coupling of liquid chromatography (HPLC) with MS detection and is especially suited for analyzing polar water soluble compounds. ESI is a "soft" technique producing primarily molecular ions (or adducts). This combination of a relatively low resolution separation with abundant molecular ions is tailor made for multiple MS techniques (MS/MS). MS/MS provides selectivity and enhanced signal to noise ratio. Another significant benefit of MS/MS is that separation of analytes from background chemical noise occurs in milliseconds, compared to the typical 40 minute HPLC run. Consequently, the HPLC can be viewed as a sample introduction system for MS, instead of MS being a chromatographic detector. Application of MS/MS techniques to surfactants and other consumer product ingredients will be used to illustrate the added value of MS/MS techniques in combination with solid phase micro extraction for estimation of bioavailability, short column LC, and HPLC. A future look at coupling high resolution liquid phase separations with MS and MS/MS is also provided.

PWA045 Micro Method for CVAAS Mercury Analysis of Tissue Samples. Steevens, J.A.*, Allgood, J.C., and Benson, W.H. The University of Mississippi, University, MS. Procedures were developed and validated for analysis of total mercury utilizing a cold vapor atomic absorption spectroscopy (CVAAS) method for use with limited amounts of tissue. Research in our laboratory has necessitated the development of a total mercury analysis for mg quantities of tissue from aquatic invertebrates such as *Hyalella azteca*. The method detection limit (MDL) was determined following the procedures outlined in 40 CFR part 136. Seven replicates of clean tissues were prepared and spiked with 0.5 ng/g methyl mercury. Replicates were analyzed and the MDL was calculated to be <0.10 ng/g. 25 to 50 mg of tissue was prepared by acid digestion, oxidation with potassium permanganate and potassium A-20 atomic absorption spectrophotometer. Additional experimental data was collected from three different groups of *Hyalella azteca* (n = 20) exposed to three levels of methyl mercury (10, 50, and 100 ppb). The three groups were analyzed and tissue residue concentrations obtained for each exposure concentration. In addition, water samples were collected and analyzed to confirm the exposure concentration for each group. The micro method outlined has several advantages over the standard CVAAS method used for mercury analysis. The first is the ability to analyze limited amounts of tissue. The standard method requires 2.0 to 5.0 g of tissue for analysis compared to only needing 25 to 50 mg of tissue using the micro method. Secondly, mercury analysis of tissues utilizing the described micro method offers a more economical and efficient alternative to the traditional digestion and preparation method utilizing less reagent and time, but at the same time is as effective as the traditional method.

PWA046 Rapid Iron Metal Reactor Treatment for the Removal of Ortho-Phosphorous from High Flow – Low Concentration Effluents. Steinhoff P. and Möller, G.*., University of Idaho, Moscow, ID. Effluent discharges on the scale of 1×10^6 G/d often represent undesirable phosphate mass loading to surface waters even for relatively low (<1 mg/L) phosphate concentrations. The use of rapid-flow, iron metal reactors for treatment of aquaculture effluent and potato food-processing effluent is examined in a pilot bench scale study. Iron metal is unstable in water and spontaneously oxidizes to form ferrous and ferric iron while creating a highly reducing environment (E^0 = -447 mV). The ferric iron shed by this process allows the formation of insoluble iron phosphate, $FePO_4$. Using iron granules as a stationary phase rapid flow system, we demonstrate near complete (98%) removal of the ortho-phosphate mass load in a variety of aquaculture test effluents and (>99%) removal of the ortho-phosphate mass load in two potato waste test effluents. The concomitant removal of nitrate in aquaculture effluent through chemical reduction to ammonia and subsequent air stripping is demonstrated at 6 - 45% in a non-optimized, rapid-flow reactor configuration. In the potato food processing effluent, the initial solution pH of 4.3 was increased post-treatment to 7.1. Although not examined in this study, other work has shown that ferric iron will allow the removal of dissolved solids such as humates by chelation and flocculation. Additional work has demonstrated the removal of heavy metals of concern from industrial waters. This pilot study supports further research and engineering study into the use of iron metal, rapid-flow reactor treatment for nutrient load reduction in high flow – low concentration effluents. A preliminary cost analysis indicates iron metal reagent consumption at $7.50 per day to treat a 1×10^6 G/d discharge containing 1 mg/L ortho-phosphate.

PWA047 Round Robin Study Results: Sediment Analysis Using a Bioconcentration Screening Method. Sheedy*, B.R., U.S. EPA, Duluth, MN; Silva-Wilkinson, R.A., Great Lakes Environmental Center, Traverse City, MI; Burkhard, L.P., U.S. EPA, Duluth, MN; DeGraeve, G.M., Great Lakes Environmental Center, Traverse City, MI; Morrow, W.J., U.S. EPA, Washington DC. The U.S. Environmental Protection Agency (EPA) has developed non-chemical-specific analytical procedures to be used to screen samples of water (effluent) tissue, and sediment for non-polar, acid-stable bioconcentratable/bioaccumulative chemicals using gas chromatography/mass spectroscopy with library searching. These methods are associated with the draft guidance, Assessment and Control of Bioconcentratable Chemicals in Surface Waters (Fed. Reg. Vol. 56, No. 61, 1991). The application of these methods results in the isolation and tentative identification (classification) of non-polar organic chemicals, including those which are not routinely monitored and potentially could form residues in aquatic organisms. A round-robin study, involving fifteen laboratories from academia, government and industry, was completed to assess the inter- and intra-laboratory variability of the analytical methods. Participants were provided with samples of sediment, effluent and tissue which they prepared and analyzed according to the methods. The reported data for the sample components and for spiked chemicals were compared and statistically analyzed. The results of the sediment evaluation are presented, characterizing the performance of the method and the similarity of the results between laboratories.

PWA048 The Effects of Adsorption on the Reusability of Tedlar Air Sampling Bags. McGarvey, L.J.* and Shorten, C.V. West Chester University, West Chester, PA. We examined the adsorption and desorption behavior of six different compounds stored in Tedlar® bags. Losses of the analytes placed at initial concentrations of ~90 ppm were observed over three-week periods. Triplicate Tedlar® bags were filled from a common liquid mixture volatilized into three distinct gas samples and each bag was sampled at predetermined intervals. Concentrations of target compounds in the gas phase were determined by capillary gas chromatography. A first-order decay model was fitted to the data using non-linear regression, and measured adsorption rate coefficients for the six compounds ranged from zero for methyl tert-butyl ether to 0.09 day^{-1} for methanol. Losses of allyl alcohol, styrene, ethylbenzene and propylene oxide demonstrated rate coefficients ranging from 0.01 to 0.03 day^{-1}. Losses of the compounds were attributed to adsorption since all other potential losses were controlled. At the end of the study the bags were cleaned using nitrogen-flushing and heating steps with reassessment after each step to determine the effectiveness of the procedure. A one-way ANOVA was used to evaluate difference in residual levels before, during and after the cleaning steps. Flushing of each bag five times with nitrogen was adequate to remove residual methyl tert-butyl ether, allyl alcohol, and propylene oxide. Combinations of flushing and heating failed to remove all the styrene and ethylbenzene, as concentrations above detection limits were observed in samples collected after each cleaning step. Neither flushing nor heating removed any of the adsorbed methanol and adsorption appeared to be irreversible. We suspect that losses may be attributable to chemisorption to the reactive fluorine groups of the bag material.

PWA049 Identification and Quantification of a Wide Range of Polycyclic Aromatic Hydrocarbons in a Coal Tar Standard Reference Material. Poster, D.L.*, Lopez de Alda, M.J., Wise, S.A., National Institute of Standards and Technology, Gaithersburg, MD. SRM 1597, Complex Mixture of Polycyclic Aromatic Hydrocarbons from Coal Tar, is a Standard Reference Material that is a natural, complex, combustion-related mixture of PAHs isolated from coal tar that was issued in 1987. Certified concentrations, based on the agreement of results from both gas chromatography and liquid chromatography, are currently available for 12 PAHs and noncertified concentrations are available for an additional 18 compounds. This material has recently been reanalyzed for additional PAHs, including many alkyl-substituted PAHs such as dimethyl phenanthrenes, methyl fluoranthenes, and methyl pyrenes, in an effort to expand the usefulness of the material. In particular, the analytical method deployed in this work included effective sample clean-up and the selection of specific stationary phases to accomplish unique separations of individual PAHs and alkyl-substituted PAHs. Clean-up involved the use of aminopropylsilane solid phase extraction followed by normal-phase liquid chromatographic isolation of the PAHs and alkyl-substituted PAHs based on the number of aromatic carbons. The aromatic ring fractions were then separated by high resolution gas chromatography using two different stationary phases with different selectivities (5% phenyl-methylpolysiloxane and 50 % phenyl-methylpolysiloxane) and analyzed using mass spectrometry. Measurements of PAHs in SRM 1597 will be presented with an emphasis on the approach used for the determination of the individual PAHs and alkyl-substituted PAHs. In addition, the relative concentrations and distributions of alkyl-substituted PAHs will be discussed.

PWA050 A New Method for the Measurement of Airborne Formaldehyde Using 3,5-bis(trifluoromethylphenyl)hydrazine. Marsella, A.M.*, Mabury, S.A., Purdham, J.T. and Ellis, D.A., University of Toronto, Toronto, ON. Aldehydes are ubiquitous indoor and outdoor airborne contaminants, originating from a variety of sources. While numerous methods are currently employed in the measurement of airborne aldehydes, environmental analysis most often makes use of C-18 or silica solid phase extraction cartridges impregnated with 2,4-dinitrophenylhydrazine (DNPH) with analysis by HPLC-UV. Unfortunately, the DNPH derivatizing agent is not well suited for analysis by GC, since the nitro moieties greatly inhibit volatility. A new method is described for monitoring formaldehyde using C-18 solid phase extraction cartridges impregnated with 3,5-bis(trifluoromethylphenyl)hydrazine (TFMPH). This method offers several advantages over the DNPH method, including the use of GC-ECD or GC-NPD in the analysis. The six fluorines provide sensitive analysis by ECD. The sensitivity and selectivity of the ECD for TFMPH derivatives is a significant improvement over the UV detection utilized by the DNPH method. Also, GC analysis provides greater peak resolution than HPLC, possibly useful in analyzing complex samples. An additional advantage is the potential for using F^{19} NMR in the analysis of TFMPH-aldehyde hydrazone derivatives, a possibility which is unique to TFMPH. Synthesis of a pure hydrazone standard (81% yield) is also described, as well as the prospect of applying the TFMPH method to the analysis of other aldehydes. All peak identities were confirmed using GC-MS.

PWA051 Effects of Sample Preparation on the Concentration of Organic Carbon, Hydrogen, Nitrogen, Sulfur and Oxygen in Marine Sediments. Ryba, S.A., Burgess, R.M., U.S. EPA, Atlantic Ecology Division, Narragansett, RI. Determining the elemental composition of marine sediment provides useful information for the study of geochemical processes, including biogeochemical cycling and contaminant partitioning. It is common practice to acidify marine sediment samples prior to elemental analysis for carbon (C) and nitrogen (N). To date, the effects of acidification on the concentrations of hydrogen (H), nitrogen (N), sulfur (S) and oxygen (O) in marine sediment have not been explicitly addressed. Acidification may result in the contamination or alteration of the sediment samples resulting in experimental artifacts. These artifacts may affect the validity of H/C, C/N and O/C ratios commonly used in geochemical studies. The objective of this study was to quantify how various preparation techniques affect the measured concentrations of C, H, N, S, and O in marine sediments. Five sediments from Long Island Sound and Narragansett Bay were evaluated in four treatments: whole (unmanipulated), HCl vapor acidification, HCl acidification by direct addition, and dry combustion. An increase in the severity of acidification effectuated a decrease in the concentrations of C and O. As the severity of acidification was increased, an elevation in the concentration of H was observed, which suggests the acid contaminated the sample. Neither nitrogen nor sulfur concentrations were significantly affected by the various acidification methods. Combustion significantly reduced all elemental concentrations compared to the whole sample. Our study supports previous work indicating direct addition acidification as the most accurate method for organic carbon and nitrogen measurement. Analysis of whole sediment is a more reliable method for the determination of hydrogen and sulfur content.

PWA052 Quantitative Determination of Bisphenol-a in Environmental Samples by Cool On-column Injection - Gas Chromatography - Mass Spectrometry. Markham, D.A., McNett, D.A., Birk, K.H., Klecka, G.M., Bartels, M.J., West, R.J., The Dow Chemical Company, Midland, MI; Staples, C.A., Assessment Technologies Inc., Fairfax, VA. Bisphenol-A (BPA) is used in industry for a variety of applications including the production of polymers and resins. To support the evaluation of environmental exposures to BPA, a sensitive and selective method was developed for analysis of the compound in environmental samples such as river waters. A review of analytical methods available at the time of the study did not reveal simple and rugged methods that provided simultaneous quantitation and confirmation of BPA at the desired detection limit of 1 ug/L. A method was developed that employs a stable isotope internal standard (D_8-BPA), liquid extraction, concentration, and analysis by cool-on-column injection, gas chromatography and detection by electron impact mass spectrometry (COC-GC-MS). The analysis involves detection of the M^+ ion and the $[M - CH_3]^+$ ion with quantitation based on the $[M - CH_3]^+$ ion and BPA confirmation based on the $M^+ / [M - CH_3]^+$ ion ratio. The quantitation limit, with confirmation, for BPA was determined to be 1 µg BPA/L, and the average relative recovery of BPA from river water was 102% ± 7% (1 to 20 µg/L). The above method, combined with appropriate matrix standards, controls and spikes was shown to avoid false positives inherent in less stringent methods commonly employed to monitor BPA in the environment.

PWA053 Enhanced bioavailability of phenanthrene to *Eisenia fetida* through the use of nonionic surfactant in artificial soil. LeBlanc, S.C.*; Dembowski, A.; Lanno, R.P., Oklahoma State University Dept. of Zoology, Stillwater, OK. Artificial soil toxicity tests were conducted to evaluate the bioaccumulation and toxicity of the polycyclic aromatic hydrocarbon (PAH) phenanthrene in soil to the earthworm *Eisenia fetida* with and without the addition of nonionic surfactant. Toxicity of phenanthrene measured by incipient lethal levels (ILLs) was increased by the presence of nonionic surfactant. Time to first mortality was the same for both the surfactant/PAH and PAH exposed earthworms. however, mortality of surfactant/PAH exposed earthworms extended beyond the mortality period for earthworms exposed to phenanthrene alone. This period of increased mortality suggests an increased solubility of phenanthrene in the sol solution due to the presence of surfactant. Increased solubility led to increased bioavailability resulting in greater mortality for surfactant/PAH exposed earthworms. Phenanthrene levels in soil and earthworm tissue will be measured and discussed in light of the mortality data.

PWA054 Assessment of the Toxicity and Bioavailability of Mixtures of Chlorophenols to *Eisenia fetida* in Soil. B. Knight*; Lanno, R., Dept. of Zoology, Oklahoma State University,Stillwater, OK. The toxicity of mixtures of chlorophenols to the earthworm Eisenia fetida have been assessed using toxic units. Incipient lethal levels (ILLs) and toxicity half-lives ($t_{1/2}$) have been determined for two mixtures of chlorophenols, However, critical body residues (CBRs) may be a better tool for determining the toxicity and bioavailability of mixtures in soil systems. Modifying factors such as pH and soil organic matter can drastically alter bioavailability and toxicity. *Eisenia fetida* were exposed to para-monochlorophenol (MCP), 2,4,5-trichlorophenol (TCP) and pentachlorophenol (PCP) during 14-day toxicity tests with artificial soil. Toxicity curves were established to determine ILLs and $t_{1/2}$s for each chemical. The ILLs for MCP, TCP, and PCP are 164, 65 and 16 mg kg^{-1}, respectively. The $t_{1/2}$s are 39, 47 and 420 hours, respectively. The ILL and $t_{1/2}$ for the mixture tests with MCP, TCP, and PCP were 1.12 toxic units and 40 hours, respectively and effects are additive. The ILL and $t_{1/2}$ for the TCP and PCP mixture test were 0.58 toxic units and 177 hours, respectively and the effects appear to be synergistic. Earthworms from all individual and mixture toxicity tests will be analyzed to determine the CBR at lethality for these chemicals. The interaction of chemicals in mixtures is poorly understood.

PWA055 Effects of Chlorpyrifos, Dieldrin, and Permethrin, on the Host-Parasite Relationship in the Terrestrial Arthropod, *Acheta domesticus*. Overmyer, J.*, Noblet, R., Klaine, S., The Institute of Wildlife and Environmental Toxicology, Department of Environmental Toxicology, Pendleton, SC. Insects are naturally subjected to a variety of biotic stresses including pathogens and parasites during the juvenile and adult stages of their life-cycle. Therefore, a healthy and efficient immune system is necessary to ward off these potential invaders to ensure survival and the ability to reproduce. In this study, the cricket, *Acheta domesticus*, was used to determine the effects of chlorpyrifos, permethrin, and dieldrin on the host-parasite relationship. Sublethal doses equivalent to 1, 25, and 50% of the calculated LD_{50} were topically applied to the abdomen of adult female crickets. After treatment with the pesticide, insects were subjected to the parasitic nematode, *Steinernema carpocapsae*, for 24 hrs at concentrations equivalent to the LD_{10}. Nematode application was conducted at two different time intervals, one at 1 day post-pesticide exposure and one at 7 days post-exposure. Results of the host resistance test showed that *A. domesticus* was more susceptible to parasitic attack by *S. carpocapsae* at 1 day post-exposure to chlorpyrifos at 25 and 50% of the LD_{50}, permethrin at 50% of the LD_{50}, and dieldrin at 25% of the LD_{50}. The synergistic effect of the pesticide/nematode combination was not observed at 7 days post-pesticide exposure.

PWA056 Emergence of Darkwinged Fungus Gnats from Packaged Commercial Soilless Greenhouse Growing Media. Taylor, M. D.*, Department of Environmental Toxicology, Pendleton, SC; Willey, R. D., Department of Entomology, Clemson, SC; Noblet, R., Department of Environmental Toxicology, Pendleton, SC. Commercially available greenhouse soilless growing media have been generally considered to be "sterile" soils. We tested eleven brands of soilless media obtained from three soil manufacturers for the presence of viable darkwinged fungus gnat (*Bradysia* spp.) eggs by placing pots of soilless media in fly-proof cages. Darkwinged fungus gnats adults (*Bradysia impatiens* Johannsen) emerged from nine brands and from each bag of two brands where we tested multiple bags of media. The bags of soilless media were stored, double bagged, in black plastic trash bags. The brands of soilless media tested included peat moss-only and combination pine bark and peat moss soils. Adults emerged from both types of soil formulations. The heavy infestation found in two media types that are mostly composted pine bark strongly suggests multiple sources of darkwinged fungus gnat eggs. Our findings have serious pest management implications for greenhouse operators. Year-round producers are unknowingly introducing this pest into their greenhouse with each crop they plant. Seasonal producers, who depend on "down times" between major crops to eradicate pests, may actually synchronize development in this pest leading to population explosions. Integrated pest management practices will need to be modified to account for both continuous introduction and synchronization of this pest.

PWA057 The Influence of Larval Age, Instar, Sex, and Colony Generation on the Response of *Bradysia impatiens* (Johan.) (Diptera: Sciaridae) Larvae to *Bacillus thuringiensis* Var. *israelensis* (H-14) (De Barjac) Utilizing a New Bioassay. Willey, R. D.*, Department of Entomology, Clemson, SC; Taylor, M. D., Department of Environmental Toxicology, Pendleton, SC; Smith, E. E., The Institute of Environmental and Human Health, Texas Tech University, Lubbock, TX; Noblet, R., Department of Environmental Toxicology, Pendleton, SC. Laboratory trials were conducted with the entomopathogenic bacterium, *Bacillus thuringiensis* (Berliner) var. *israelensis* (Serotype H-14) (de Barjac) (Bti) against laboratory reared *Bradysia impatiens* (Johan.) larvae employing a new, rapid and inexpensive bioassay. Results indicated that susceptibility (the number of ITU/mg of Bti needed to kill a desired percentage of the test organisms) to Bti of 1^{st} instar larvae was 45 fold higher compared to 4^{th} instar larvae; however, sensitivity (the increment of increase in mortality for a unit increase in Bti concentration) to Bti significantly increased from 1^{st} to later instars. We found that while 2^{nd} instar larvae are not the most susceptible age, they are the instar of choice for Bti bioassay work because they are large enough to work with, show good sensitivity to Bti, have low control mortality, and may be utilized with little concern for mixing ages within the instar in a test chamber. There was no difference in susceptibility to Bti among 2^{nd} instar larvae of the monogenic species *B. impatiens* (Johan.) based on larval sex (p=0.41). Comparisons of established colonies' susceptibility to Bti with susceptibility of first generation larvae from collected "wild-type" adults demonstrated that long term laboratory culturing does not appear to increase or decrease susceptibility to Bti. Carefully maintained cultures do not weaken from disease or inbreeding over multiple generations. *B. impatiens* (Johan.) exhibits a range of susceptibility to Bti that may be related to the origin of the colony being tested (strain variability from genetic factors). This range of susceptibility based on colony origin suggests that bioassays should employ only larvae taken from a single colony. The 45 fold decrease in susceptibility to Bti from early 1^{st} instar to 4^{th} instar means that it is very important for greenhouse and nursery growers and mushroom producers to treat crops early in the insect developmental cycle both to achieve insect control and to reduce pesticide expenses and application costs.

PWA058 Use of Toxicity Reference Values versus Soil Quality Criteria and Guidelines in Development of Soil Ecological Data Quality Levels (EDQL). Mazur, D.J.*, U.S. EPA, Chicago, IL; Monschein, E.S., Tetra Tech EM Inc., Chicago, IL. The U.S. Environmental Protection Agency, Region 5 determined a consistent set of EDQLs was needed to avoid redundant development of toxicity benchmarks for each screening ecological risk assessment. These EDQLs are needed to identify levels of contamination for each environmental medium (air, sediment, soil, and water) at a Resource Conservation and Recovery Act (RCRA) facility whose exceedance would warrant further investigation. EDQLs are not intended to serve as cleanup or action levels. During development of EDQLs for soil, various state, federal, and international soil quality criteria and guidelines were assessed. While each soil quality criterion and guidelines were designed to protect human receptors, ecological receptors, or both, the assessment revealed significant variation in their methods of development, toxicological endpoint, and applicability to ecological receptors. Due to the significant variation in chemical-specific soil quality criterion and guidelines, EDQLs are now based entirely on toxicity reference values adjusted to specific ecological receptors and simplified food chains.

PWA059 Determining Bioavailability of Heavy Metal Contaminated Soils With and Without Phosphorus Amendments Using a Toxicokinetic Approach. Pearson, M.S.*, and Lydy, M.J., Wichita State University, Wichita, KS. Large scale mining operations in southeast Kansas were conducted from the mid 1800's through the 1950's. High concentrations of lead (Pb), zinc (Zn), and cadmium (Cd) exist throughout the Tri-State Mining Area which includes parts of Kansas, Oklahoma and Missouri. Pb, Zn, and Cd soil concentrations have been found to be as high as 66,900, 15,750, and 400 mg/kg, respectively. Because many parts of the Tri-State Mining Area have been declared a Superfund site, clean-up or some form of remediation has been mandated by USEPA. Recently, phosphorus and organic matter soil amendments have shown promise as a possible on-site remediation tool for metal contaminated soils in urban areas. We employed a toxicokinetic approach to determine if phosphorus soil amendments reduce the bioavailability of metals in contaminated soils to earthworms. We exposed earthworms (*Eisenia foetidia*) to an artificial soil with nominal concentrations of 1000 mg/kg Pb over a 16 d period, with and without phosphorus amendments. Replicate *E. foetidia* and soil samples were removed at 12, 24, 48, 96, 192, and 384 h, acid digested, and analyzed by flame atomic absorption spectroscopy. Two-way ANOVA show earthworm Pb concentrations to be significantly ($p = 0.001$) different among phosphorus amendments (with and without), and Tukey's multiple range test indicated that steady state was achieved by 96 h. Toxicokinetic parameters (k_s, k_d, and BAF's) for earthworms in soils amended with phosphorus have lower uptake rates, elimination rates, and BAF's than earthworms in soils without phosphorus amendments. Results of these bioaccumulation experiments show that soils amended with phosphorus will reduce the bioavailability of heavy metals to terrestrial organisms.

PWA060 Quantifying Behavioral Changes in the Nematode, *Caenorhabditis elegans* using Computer Tracking. †Dhawan, R.*, ‡Dusenbery, D. B.,and. †Williams, P. L.† Environmental Health Science, University of Georgia, Athens, GA, USA, and ‡ School of Biology, Georgia Institute of Technology, Atlanta, GA, USA. Behavioral changes are one of the most sensitive indicators of toxicant exposure, but can be very difficult to quantify mainly due to methodological problems. Using the free-living soil nematode, *Caenorhabditis elegans*, a computer tracking system has been developed that simultaneously measures several behavioral parameters for approximately 100 subjects on a real-time basis. Toxicant exposure are performed in an aquatic media, following the exposure duration, the worms are transferred to a thin-layered agar slab. Worms were viewed in dark-field illumination by a video camera interfaced directly to a microcomputer. Information is provided on the number of subjects moving, their individual rate of locomotion, and the frequency of change in direction. This study assess the influence of several metals on the behavior of worms. The nematodes were exposed for 24 hours with food concentrations of an individual metal in an aquatic media representing the interstitial pore water within the soil. Under identical conditions, concurrent LC50 values were determined for each metal tested. The tracking system found significant changes in several behavioral parameters (e.g., rate of locomotion, number of subjects moving) at concentrations of less than 5% of the respective LC50 value. The tracking system is straight forward in use and is a very sensitive and reliable indicator of toxicity.

PWA061 Ion Exchange Membrane Uptake and Toxicity of Heavy Metals to *Eisenia fetida* in Soil. Conder, J.M.*, Lanno, R.P., Oklahoma State University, Stillwater, OK. There is a great need for an inexpensive, reliable, and more accurate measure of bioavailable heavy metals in soil. Plant Root Simulators™ (PRS™, Western Ag Innovations, Saskatoon, SK, Canada) are DTPA-impregnated, anion exchange membranes designed to mimic metal uptake by plant roots through a direct soil exposure. Soil toxicity tests using the earthworm *Eisenia fetida* and PRS uptake in artificial soil will be conducted for Cd, Pb, and Zn, individually and as a three-metal mixture. Incipient lethal levels (time-independent mortality) for *E. fetida* exposed in artificial soil to Cd, Pb, and Zn were 2918, 3162, and 2051 mg/kg, respectively. PRS uptake will be compared to toxic responses and mixture toxicity will be assessed using the Toxic Unit method.

PWA062 Fluctuating asymmetry in terrestrial isopods as a potential indicator of hazardous metal exposure. Peters, E.L., Chicago State University, Chicago, IL. Fluctuating asymmetry (FA) in terrestrial isopods (woodlice, pillbugs) is being evaluated as a potential indicator of the presence of metal contaminants in urban residential environments. Terrestrial isopods are soil-dwelling detritivores that are distributed throughout the Northern Hemisphere, where they are abundant even in urban areas. Isopods have limited dispersal ability, are easily collected by active or passive sampling, and can be reared in the laboratory for genetic and/or behavioral toxicology studies. Isopods are also known to accumulate large amounts of hazardous metals (one or more percent of Cu, Zn, Cd, and Pb by dry mass, among the highest measured in any organism). We have noted significant increases in deviations from an expected mean difference of zero in the number of compound eye lenses (ommatidia) of the terrestrial isopod *Armadillidium vulgare* from metal-contaminated sites in south Chicago compared with isopods collected in the suburbs. All populations had high levels of Ba and Mn, and all isopod populations in the city showed higher levels of Cr, Fe, Cu, Zn, Cd than the suburban population. Be and Hg were not detectable in the suburban population, but were present in all city samples. The degree of FA was positively correlated ($r^2 = 0.89$) with the total molar concentrations of the ten metals examined (Be, Cr, Mn, Fe, Cu, Zn, Cd, Ba, Hg, and Pb), and was particularly well correlated (r^2 • 0.90)with concentrations of Fe, Cd, and Pb.

PWA063 Mapping toxic stress (expressed as PAF) of some heavy metals in Dutch soil. Klepper, O.; Traas, T.P.*; Van de Meent, D., National Institute of Public Health and the Environment (RIVM), Bilthoven, The Netherlands. In order to prioritize environmental policies it is necessary (i) to compare the toxic effects of different substances and (ii) to evaluate combined toxicity of different sites. Evaluating the ratio of environmental concentration over the no-effect concentration is of limited value, as the same ration may have widely different effects for different substances. In this study, the fraction of species exposed to a concentration above their no-observed effect concentration (NOEC) or the Potentially Affected Fraction of species (PAF) was used as a basis for comparison. Cumulative distributions of NOECs are determined from literature data. By combining maps of soil concentrations with estimates of bioavailability and estimates of natural background concentrations, the antropogenic PAF was determined for the most important heavy metals. Combination toxicity is calculated under the assumption that the individual toxicants act independently (effect additivity). The results show the utility of the approach in ranking the toxicities of substances and areas. The results also reveal the sensitivity of the model for different types of uncertainties (mainly bioavailability and species sensitivity distribution).

PWA064 Evaluating Explosives-Contaminated Soils and Compost Mixtures Using The Earthworm Avoidance Test. Jarvis, A.S.*, USAEWES, Vicksburg, MS; Inouye, L.S., AScI Corporation, Vicksburg, MS; McFarland, V.A., USAEWES, Vicksburg, MS. Contamination of soils with explosives is a common environmental problem at formerly-used defense sites, munitions plants, and military bases undergoing closure. Composting explosives-contaminated soils has emerged as one of the most popular remediation treatments due to its relatively low cost compared with alternatives. The purpose of this study was to evaluate the ability of a simple behavioral test to monitor the effectiveness of composting as a remediation technique. Reference soils were spiked with 2,4,6-trinitrotoluene (TNT), hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX), and octahydro-1,3,5,7-tetranitro-1,3,5,7 tetrazocine (HMX). LC_{50} values were determined to establish toxicity. TNT-spiked soils were tested at the LC_{10} (280 mg/kg). HMX and RDX-spiked soils were not acutely toxic and were tested at 417 mg/kg and 402 mg/kg, respectively. Additional aliquots of the TNT, RDX, and HMX-spiked soils were composted for four weeks. The composted TNT-spiked mixture was tested at the same dilution as the TNT-spiked soil. The composted RDX-spiked mixture was tested at approximately 201 mg/kg because of initial toxicity following composting. In the avoidance test, earthworms, *Eisenia foetida* were exposed for a period of four days in circular test chambers containing an artificial soil control in one hemisphere, and a test soil or compost mixture in the other. An avoidance response (100 %) was clearly demonstrated in the TNT-spiked soil while no significant differences were observed for RDX and HMX spiked soils (58.7% and 59.84%). The composted TNT-spiked mixture was avoided (72%) as was the composted RDX-spiked mixture (71 %) and composted uncontaminated soil. It appears from these results that the earthworm avoidance test is unsuitable for monitoring clean-up effectiveness of composting because the process of composting itself alters the soil in such a way that contact with it is stressful to earthworms.

PWA065 An Evaluation of Plant Uptake of Lead from Soil at a Small Arms Firing Range. DeShields, B.R.*, Harding Lawson Associates, Novato, CA; R. Toia, University of San Francisco, San Francisco, CA. The beach dunes at the former Fort Ord were historically used as small arms firing ranges, resulting in the accumulation of spent bullet fragments and elevated concentrations of metals, predominantly lead, in near-surface soils. Of particular interest at the beach firing ranges at Fort Ord are two species of buckwheat that provide habitat for an endangered butterfly. In previous studies, field measurements of buckwheat health and condition did not indicate adverse impacts on buckwheat plants onsite from exposure to lead, except in the areas of highest surface bullet cover. Additional field studies were conducted to characterize lead concentrations in onsite soils and above-ground plant tissues. A plant uptake assay was conducted to validate the findings of the field studies and to evaluate the partitioning of lead between below-ground (root) and aboveground (shoot) plant tissues. A positive correlation between soil and plant tissue lead concentrations was demonstrated. Plant uptake factors (PUF) and shoot-to-root ratios were calculated. A negative correlation between PUF and soil lead concentration was demonstrated (as soil concentration increases, the rate of plant uptake decreases). A positive correlation between shoot-to-root ratio and soil lead concentration was demonstrated (as soil concentration increases, a higher proportion of lead is taken up into the above-ground plant tissues). In addition, PUFs from firing range soils were shown to be lower than those for reference areas and soils spiked with a soluble form of lead, indicating that the form of lead present in firing range soils is not highly bioavailable.

PWA066 RATL: A reptile and amphibian ecotoxicology database. Pauli, B.D.* and Money, S.L. Canadian Wildlife Service, National Wildlife Research Centre, Hull, PQ, Canada. Due to an unexplained decline in global amphibian populations, and a decline in many reptile populations, there is intense interest in the effects of environmental contaminants on these animals. The objective of this project is to establish a searchable database on the effects of environmental contaminants on amphibians and reptiles. The Reptile and Amphibian Toxicology Literature (RATL) database contains data extracted from the primary literature. A graphical user interface allows the user to perform custom searches and generate reports. The database can be searched in a number of ways: by contaminant group, common name, trade name or CAS number; by species, genus, or higher taxonomic group; by author; or by certain toxicological effects categories including hatching success and developmental abnormalities. Combined searches are also possible. Currently, there is approximately 1300 references in the database with toxicity information entered to 1995. The database will be brought up to date and future updates are planned. A further component of this project is to analyse the collected information in terms of the comparative toxicity of compounds or classes of compounds to amphibians in relation to other aquatic organisms.

PWA067 Genetic Variation in *Rana sphenocephala* in Response to an Agricultural Chemical: Implications for Amphibian Conservation. Bridges, C.M. University of Missouri, Columbia, MO. The amount of genetic variability present may indicate a species' ability to adapt to a stressful environment and can be examined in a quantitative genetic analysis. Currently, numerous conservation efforts are being devoted to determining whether amphibian species are declining at an alarming rate, as many studies would suggest. Therefore, because amphibians are amenable to quantitative genetic analyses and because they may be declining in numbers due to changes in their environment, these organisms serve as an excellent model for examining the relationship between the amount of genetic variability present in populations and adaptability in changing environments. Using tadpoles of a single southern leopard frog (*Rana sphenocephala*) population, I examined the amount of genetic variability among full- and half-sib families with respect to their sensitivity to both lethal (time-to-death) and sublethal levels (change in activity) of the agricultural chemical, carbaryl. Analyses of both time-to-death and activity data indicates significant differences among full sib families, suggesting a great deal of variability, genetic or environmental, present in the responses to this environmental stressor. There were also significant differences among half sib families in the degree to which activity was reduced by sublethal carbaryl levels, indicating that this variability has a genetic basis.

PWA068 Effects of an insecticide on amphibian communities under different levels of biotic stress. Boone, M.D., University of Missouri, Columbia, Missouri. Widespread use of agricultural chemicals makes it increasingly likely that non-target wildlife could be exposed to contamination. Many amphibian species utilize breeding habitats nested within or around agricultural areas, therefore population and community dynamics may be influenced or altered by exposure to chemicals. The chemical used in this experiment, carbaryl, is the active ingredient in the insecticide *Sevin*. This chemical acts through acetyl-cholinesterase inhibition, and is known to affect amphibian activity and behavior. The objectives of this experiment were to determine differential susceptibility to carbaryl among species, the effect of the chemical under different biotic conditions, and the indirect effects of the chemical. To do this, I used three species of anurans in outdoor cattle tanks: *Hyla versicolor* (gray treefrog), *Bufo woodhousii* (toad) and *Rana clamitans* (green frog). Twelve experimental treatments were manipulated for density (high or low), predator (presence or absence of *(Notophthalmus viridescens)*, and chemical level (0, 3.5, or 7.0 mg/l of carbaryl). Results showed that the chemical significantly affected size at metamorphosis in *H. versicolor*. A predator by chemical effect in *H. versicolor* was significant; in the presence of the predator, tadpoles at high chemical levels had a larger size at metamorphosis than other classes. Chemical effects on survival at metamorphosis were significant in *B. woodhousii* and *H. versicolor*; higher chemical levels significantly reduced survival. The chemical interacts with biotic factors in ways that would not be predictable from two-factor experiments. Chemical by density interactions were significant in *B. woodhousii* and *H. versicolor*. This experiment does indicate differential susceptibility among species and noted that differences in biotic conditions affect the potency of the chemical.

PWA069 Amphibian and Reptilian Toxicology: History and Research Needs. Sheffield, S.R., Dancik, J., Casey, R., Huddleston, G.M., Smink, J., Eversen, M., Clemson University, Pendleton, SC. Amphibians and reptiles as a group have been largely neglected in toxicological studies. Our objectives of this paper are to outline the history of amphibian and reptilian toxicological research, including research currently being conducted at Clemson University, and to suggest regulatory and research needs for these taxa. Amphibians have enjoyed some attention in recent years due to use of the FETAX assay and the press they have received on malformations and population declines. Reptiles have been largely overlooked, with the exception of some attention being paid to snapping turtles and Lake Apopka alligators. One reason for this is that it is assumed that regulations aimed at protecting fish, birds and mammals also protect amphibian and reptilian species. There has been little work done in the way of comparative vertebrate toxicology, and current evidence points to the possibility that amphibians and reptilian species may not be protected, making this assumption invalid. To address this, we suggest that model amphibian and reptilian species be included in FIFRA testing as once proposed, to include at least one terrestrial and one aquatic species of each taxa. Future work involving amphibian and reptilian toxicology should include (1) increasing the amphibian and reptilian toxicological database so that we can begin to reliably include these taxa in ecological risk assessments, (2) further work on the role of contaminants in amphibian and reptilian malformations and population declines, (3) more focus on comparative toxicology including amphibian and reptilian species, (4) more basic research using unexposed amphibian and reptilian species to examine biochemical and molecular processes that could serve as useful biomarkers, and (5) more research on amphibian and reptilian species that are regularly consumed by humans, particularly long-lived species such as turtles and alligators.

PWA070 **Effects of Sublethal Exposure of the OP Insecticide Chlorpyrifos on Fitness Parameters, Thyroid Hormone Levels, Behavior, and Immunocompetence of Larval Amphibians.** Day, B.S.*, Clemson University, Pendleton, SC; Sheffield, S.R., Clemson University, Pendleton, SC. Three species of amphibian, the leopard frog (*Rana pipiens*), gray tree frog (*Hyla chrysoscelis*), and tiger salamander (*Ambystoma tigrinum*), are being tested for possible effects of sublethal exposure to the organophosphate (OP) insecticide chlorpyrifos. The LC50 for 7-day old *R. pipiens* was found to be approximately 1ppm; thus, concentrations of 0.125, 0.25, and 0.5 ppm plus control are being used. Pulsed exposures to Lorsban 4E (twice for 48 hrs each at a two-week interval starting with 7-day old tadpoles) simulate realistic field conditions. Tests for each species run through metamorphosis. Larvae are monitored daily for mortality, developmental abnormalities, and aberrant behaviors. Aberrant behaviors, including twisting, aberrant swimming, and slowed response to stimuli, and developmental abnormalities, such as tail kinking were seen at concentrations as low as 0.25 ppm. At metamorphosis, growth metrics, T3/T4 levels, histopathology of the gonads, and immunocompetence are being conducted. The three species tested represent different ecological niches for amphibians; thus the data will be useful in drawing inferences about the potential impacts of a widely used OP insecticide on amphibian populations.

PWA071 **The Effect of Water Hardness and Humic Acid on the teratogenicity and toxicity of Atrazine using FETAX.** Napier, J.D.*, Scheuerman, P.R., Pyles, R.A., East Tennessee State University, Johnson City, TN. The effect of water hardness and humic acid on the teratogenic potential of atrazine was evaluated using FETAX (frog embryo teratogenic assay - *Xenopus*). This study was designed to determine the adverse effects caused by variable water hardness for atrazine with and without humic acid. Artificial natural water at three hardness levels was evaluated. Dose response data from these assays were evaluated using linear regression and PROBIT analysis. Bioassays in soft water (22 mg/l as $CaCO_3$) yielded an EC50 and LC50 of 0.03 and 204 mg/l, respectively, and a teratogenic index (TI) of 6,800. The laboratory buffered water (FETAX solution) at a hardness of 46 mg/l as $CaCO_3$ resulted in an EC50 and LC50 of 6.79 and 184 mg/l, respectively (TI = 27.1). Hard water (105 mg/l as $CaCO_3$) resulted in a higher EC50 of 11.01 mg/l and a lower LC50 value of 44.8 mg/l (TI = 4.07). These results suggest that water hardness is an important factor influencing the teratogenicity and toxicity of atrazine. Increasing water hardness shifts the effects of atrazine from teratogenic to toxic. This shift in effect may be due to permeability changes in embryonic tissues due to the increasing calcium or other ion concentrations in the test solutions. Humic acid addition to hard water resulted in an EC50 of 10.4 and LC50 of 124 mg/l as atrazine (TI = 11.9). The data suggest that humic acid provides protection against toxicity of atrazine to amphibians but does not affect the teratogenicity. In areas where water hardness is low, atrazine may pose a significant ecological health threat at environmentally relevant concentrations.

PWA072 **Estimating and Testing Bioconcentration Factors.** Bailer, A.J.*, Venis, K.J. and Walker, S.E. Center for Environmental Toxicology and Statistics, Dept. of Math. & Stat. (AJB, KJV), Dept. of Zoology (SEW), Miami University, Oxford, Ohio USA. Bioconcentration factors (BCFs) are commonly constructed to represent the equilibrium concentration of a substance in an organism relative to environmental concentrations of this same substance. The BCF is derived from parameters estimated in uptake-elimination experiments. A standard error and confidence interval for BCFs is presented. A delta method was employed to approximate the standard error. In addition, a statistical method for formally comparing the BCFs derived under two or more experimental conditions is discussed. These methods are illustrated using data from a study of fluoranthene-exposed tadpoles.

PWA073 **Comparative developmental toxicity of all-*trans* retinoic acid in limb bud stage *Xenopus laevis* and *Rana pipiens* tadpoles.** Degitz, S.J., Kosian, P.A. Makynen, E.A., Holcombe, G.W., Jensen, K.M., Ankley, G.T., US EPA, Duluth MN. Recently, high frequencies of malformations have been reported in amphibians across the United States. The most striking and commonly reported malformations are anomalies of the hind limb. The phenotype of the malformations has led to speculation that dysmorphogenesis is induced by retinoid-like compounds. The observed malformations have occurred in a number of native anurans, including the northern leopard frog *Rana pipiens*. However, because they are readily cultured in the lab, *Xenopus laevis* would be very useful as a model organism to study the risk of chemical dysmorphogenesis in native amphibians. Before this can be done, comparison of the response of *X. laevis* to retinoids with the response observed in native species is necessary. Previous studies have documented that retinoids can induce hind limb anomalies in *X. laevis*. The objective of this study was to determine if retinoids could also induce hind limb dysmorphogenesis in *R. pipiens*. *R. pipiens* tadpoles (stage 27) were exposed to concentrations of all-*trans* retinoic acid (RA) ranging from 250 ng/ml to 6500 ng/ml for 24 hours. Subsets of tadpoles were collected at 4, 12 and 24 for analysis of RA metabolism. Additional tadpoles were held until metamorphosis, stained with alcian blue and alizarin red, and hind limbs were evaluated. RA concentration of 1500 ng/ml and 2000 ng/ml resulted in 50% and 85% mortality, respectively, within 48 h. Concentrations of 3500 ng/ml and above caused 100% lethality by the end of the exposure. RA treatment (2000 ng/ml) also resulted in dysmorphogenesis of the hind limb. These results show that a pulse dose of RA at high concentration will produce dysmorphogenesis of the hind limb in *R. pipiens* as in *X. laevis*. Further studies will establish the specific dose response relationships for these two species and extend species comparison to include biomarkers of retinoid response.

PWA074 **Adapted amphibian test demonstrates long-term effects of early exposure to PCB126 on *Xenopus laevis* not visible in an early life stage test (FETAX).** Gutleb, A.C., Appelman, J., Bronkhorst, M., van den Berg, J.H.J., Murk, A.J.* Wageningen Agricultural University, NL. The classical FETAX assay is based on the principle that early life stages often are most vulnerable for toxic compounds. It is possible, however, that subtle hormonal effects are not visible within 96-hours, but result in endocrine disturbances in a later life stage. For this reason we developed a so-called prolonged-FETAX assay, in which delayed effects of former exposure become visible. In the prolonged FETAX exposure also is for a 96 hours period but after this the larvae are kept in bigger aquaria without further exposure. We compared the effects of PCB126 on *Xenopus laevis* in the standard-, and in our prolonged FETAX assay. PCB126 was dosed in a concentration range of respectively 17.1 pmol-15.5 µmol/ml and 7.7 pmol, 0.64 nmol and 6.4 µmol/ml. PCB126 had no effect on the rate of malformations, growth and development in the FETAX assay. In the prolonged-FETAX assay five days after the last PCB126 exposure mortality increased sharply in the highest dose group (6.4 µmol/ml PCB126) and 95 of the 100 tadpoles died over the whole experimental period with most animals showing swimming disorders, eye malformations and severe oedema. In the groups exposed to 0.64 nmol/ml and 7.7 pmol/ml PCB126 a steadily increased mortality resulted in 43 respectively 21 dead animals showing the same type of malformations. A dose-related increase in the rate of malformations was found in the PCB-exposed groups, with 93.2% of dead animals showing malformations in the 6.4 µmol/ml PCB126 group.

PWA074A **Effects of 2,4,6-trinitrotoluene in a holistic environmental exposure regime to a terrestrial salamander: *Ambystoma tigrinum*.** Johnson, M.S.*, U.S. Army Center for Health Promotion and Preventive Medicine, Aberdeen Proving Ground, MD; Holladay, S. D., Virginia Polytechnic Institute, Blacksburg, VA; Lippenholz, K. L., Oak

Ridge Institute for Science and Education, Oak Ridge, TN; Jenkins, J. L., National Aquarium at Baltimore, Baltimore, MD. 2,4,6-Trinitrotoluene (TNT) is a defense-related environmental contaminant present at high concentrations in soil at some military installations. Tiger Salamanders (*Ambystoma tigrinum*, Family: Ambystomidae) were exposed to TNT in a soil matrix and fed earthworms which had also been exposed to TNT via contaminated soil. Such exposure was previously shown to result in significant accumulation of both TNT and TNT metabolites by salamanders. Following 14 days of combined oral and dermal exposures, salamanders were evaluated for signs of toxicity. Control and TNT-exposed salamanders gained weight ($p < 0.025$). In addition, organ to body weight ratios (kidney, liver and spleen) were not affected by treatment. The function of splenic phagocytic cells was evaluated, since these cells have been found to be sensitive to certain environmental chemical exposures. Neither the chemiluminescence response (H_2O_2 production) nor the phagocytic capacity of such cells were different between controls and treatment. In like manner, no change was seen in the peripheral hematological parameters investigated. Histopathological evaluations were inconclusive, yet the liver revealed a high concentration of heavily pigmented, iron-rich phagocytes (melanomacrophages). Too few treatment representatives were available for an accurate characterization via image density to be made. This investigation presents a realistic approach and preliminary data for investigating the effects from xenobiotic exposure in a soil matrix for a terrestrial vertebrate.

PWA075 Behaviour of Silver in the Presence of Iron Monosulphide (FeS) under Oxidizing Conditions. Manolopoulos, H.*, McMaster University, Hamilton, ON, Canada; Kramer, J.R., McMaster University, Hamilton, ON, Canada. Silver ion (Ag^+), a type-B metal cation, will bind strongly to sulphide in anoxic sediments. However, processes such as resuspension can expose sediments to dissolved oxygen resulting in the oxidation of sulphide and sulphur-containing compounds. The fate of silver-sulphur compounds in sediments under oxic conditions has not been characterized. The main objective of this study was to examine the effect of molecular oxygen (O_2) on silver bound to iron monosulphide (FeS), a common constituent of anoxic sediments. In addition, we wanted to examine the role of thiol ligands (3-mercaptopropanoic acid and cysteine) in complexing and stabilizing silver ion under both oxic and anoxic conditions. The system was kept anoxic for the first 24 h after which it was openly exposed to the atmosphere and allowed to saturate with oxygen. Under anoxic conditions, FeS scavenged silver from solution while complexation with thiol ligands increased dissolved silver concentrations in the presence of FeS. FeS was rapidly oxidized as indicated by the precipitation of orange iron(III) hydroxides, while dissolved oxygen concentrations reached saturated levels within 25 h. Dissolved silver concentrations increased over time in both the 10 kDa and 0.45 µm filtered fractions as a consequence of oxidation processes.

PWA076 Determination of Silver Complexation in Wastewater Effluent and Surface Waters. Adams, N.W.H.* and Kramer, J.R., McMaster University, Hamilton, ON, Canada. A competitive ligand equilibration/solvent extraction CLE/SE) technique was used to examine silver complexation in the particulate (> 0.45 µm), colloidal (0.45 µm - 10 KDalton) and dissolved (< 10 KDalton) phases in wastewater effluent and surface waters. Additions of silver at near ambient levels (0.1-10 nM), followed by CLE/SE, allowed the determination of complexation sites with large stability constants, responsible for silver binding. Findings from this study are discussed in relation to previously collected data on silver and sulfide concentrations in the particulate, colloidal and dissolved phases in wastewater effluent and surface waters. The combined results of these studies suggest that silver is complexed to S(-II), even in oxygen saturated waters. The CLE/SE technique was also used to examine the long-term stability of these silver complexes with respect to shifts in pH, exposure to oxygen and photochemical decomposition. As well, the contribution of inorganic and organic ligands to silver binding was assessed through comparison of UV irradiated and non-irradiated samples.

PWA077 Aggregation and ligand exchange of silver(I) thiolates: an NMR study of D-penicillaminato-silver. Bell, R.A. and Bennett, S.E., Department of Chemistry, McMaster University, Hamilton, ON, Canada. Silver(I) thiolates exist in aqueous solution as short polymers of interlocking, and interconverting, chains of -S(R)-Ag-S(R)-Ag-zig-zag units. We have used proton and carbon-13 NMR spectroscopy to study the aggregation and ligand exchange of D-penicillaminato-silver in solution (pH 10). The ^1H NMR line widths of the C-H and two CH_3 groups in D-penicillamine itself were increased by a factor of four in the spectrum of the silver complex. Measurement of the spin-lattice relaxation times (T_1) of the C-H and two CH_3 carbons for D-penicillamine and the silver complex showed a ten-fold decrease for the complex. This is consistent with an increase in molecular mass of species in solution in proceeding from ligand to silver complex. Heating a solution of the complex showed a decrease in line widths, arising from the anticipated decrease in molecular aggregation. However, contrary to expectation, decreasing the concentration from 0.1M to 0.001M showed an *increase* in line widths by a factor of seven. This may be accounted for by a ligand exchange process; that is, some protons have environments inside an aggregate and others outside an aggregate, and the exchange process is partially averaging their environments. When an additional equivalent of D-penicillamine was added to a solution of silver complex, the ^1H and ^{13}C NMR spectra showed only a single species; that is, rapid ligand exchange is occurring. In the carbon spectrum, the shifts of the C3-S carbon may be used to estimate a minimum ligand exchange rate. Addition of dilute HS^- to solutions of the silver complex leads to formation of colloidal Ag_2S.

PWA078 X-ray Absorption Spectroscopy of Silver Compounds. Anderson, Paul R.*, Illinois Institute of Technology; Chicago, IL; Bunker, Grant B., Illinois Institute of Technology, Chicago, IL. X-ray absorption spectroscopy (XAS) can provide information on the nearest-neighbor and next-neighbor interactions of most elements in the periodic table. XAS is an important tool in environmental chemistry because it describes the electronic environment of elements in both crystalline and poorly crystalline solids such as soil clay minerals, and it provides information on the coordination environment of soluble and surface sorbed species. XAS measurements of silver are somewhat problematic because the K-absorption edge for silver is a relatively high 25.5 KeV. We will report results from XAS measurements taken at the Advanced Photon Source (APS) at Argonne National Laboratory. The APS has higher energy characteristics and a high photon flux that provide a good signal-to-noise ratio and make it possible to examine samples with relatively low silver concentrations. We examined a basis set of reference silver compounds including Ag_2O, Ag_2S, $AgNO_3$ solution, and Ag^+ adsorbed to hydrated ferric oxide. These spectra were used to describe silver speciation in a photoprocessing silver recovery precipitate, and in a model biological waste treatment sludge. Although we find silver coordinated with sulfur in both sludges, the average bond lengths are different.

PWA079 The Effect of Dissolved Organic Carbon on Silver Toxicity to *Ceriodaphnia dubia*. Bielmyer, G.B., Clemson University, Pendleton, SC, Karen, D.J., Clemson University, Pendleton, SC, Klaine, S.J., Clemson University, Pendleton, SC, Bell, R.A., McMaster University, Hamilton, Ontario, Canada. Static-renewal bioassays were conducted using *Ceriodaphnia dubia* to characterize the acute toxicity of silver bound to cysteinate and glutathionate. Both acute (mortality) and chronic (reproduction) endpoints were measured over 8 days. Total concentrations of silver cysteinate less than or equal to 6 µg/L did not exhibit either acute or chronic toxicity. Bioassay results also suggested that silver glutathionate was acutely toxic to *C. dubia* at greater than or equal to 5 µg/L as total silver; reproductive effects were present at all concentrations used (1.25-2.5ppb of Ag-GSH). Additional studies are in progress to examine the transfer of silver through a freshwater alga-daphnid food chain.

PWA080 Silver uptake and depuration in rainbow trout in the presence and absence of dissolved organic matter (DOM). Bertram, B.*, Schwartz, M., and Playle, R.C. Wilfrid Laurier University, Waterloo, Ontario, N2L 3C5. Rainbow trout (*Oncorhynchus mykiss*, ~50 g) exposed to 0.15 µ*M* silver as $AgNO_3$ in soft water quickly accumulated large amounts of Ag in their gills, plasma, and livers (within 6 h), but showed no ionoregulatory, respiratory, or toxic responses over one week. In contrast, trout exposed to 0.15 µ*M* Ag plus 25 mg $C \cdot L^{-1}$ dissolved organic carbon (DOC) did not accumulate Ag, except in their livers after one week. Depuration of accumulated Ag was studied in the presence and absence of DOM to determine if DOM can act as an additional "sink" for accelerating Ag removal from fish. There was no indication that waterborne DOM increases Ag depuration rates from the gill, plasma, or liver compartments, but Ag loading in these compartments was not very high in this preliminary study. This experiment is being repeated with higher Ag loading, to determine the influence of DOM on Ag depuration as well as on Ag uptake.

PWA081 A Metal Specific Biosensor for Quantification of Bioavailable and Bioreactive Metals. Shaw, J.R.*, Birge, W.J., Hogstrand, C., University of Kentucky, Lexington, KY and Wood, C.M., McMaster University, Hamilton, ON. The contamination of aquatic ecosystems by metals is widespread and as a consequence there is growing need for more reliable techniques to quantify not only metal bioavailability, but also the intracellular reactivity (bioreactivity) of metals. Current research in our laboratories is focusing on the development of an *in vitro* biosensor that can detect both metal bioavailability and bioreactivity. The biosensor exploits the metal responsiveness of the metallothionein (MT) promoter, the light-generation of the firefly luciferase gene product, and a newly developed technique to reconstitute a "tight" fish gill epithelium. It will employ a hybrid gene in which the rainbow trout MT-A promoter has been fused upstream of a luciferase reporter gene and cloned into a plasmid vector. The MT-luciferase gene construct will be transfected into freshly isolated rainbow trout gill cells, which will be reconstituted into a "tight" epithelium. This artificial gill is comprised of the same cell types (pavement cells, ionocytes) as in the intact gill, which orient uniformly such that apical (water) and basolateral (blood) surfaces are maintained. Raw test water will be directly introduced to the apical side of the epithelium. If inducing metals are present and enter the gill cells in a bioreactive form, the MT promoter will be activated and luciferase synthesis initiated. Thus, the metal biosensor isolates the primary target for most acute metal mediated toxicity, the gill, and will provide a test system that can quantify the bioavailability and bioreactivity of metals in natural waters under relevant water chemistry. The biosensor will be compared with other predictors/indicators of metal stress (toxicity tests, whole body metal burden in sentinel monitors, 'gill-binding' models).

PWA082 Decreasing silver concentrations in mollusks since 1986. O'Connor, T.P.* NOAA, Silver Spring, MD. NOAA Mussel Watch results for 1986 through 1996 showed decreasing trends in molluscan concentrations for chemicals whose use has been banned or severely curtailed in the United States; chlorinated hydrocarbons, butyltins, and cadmium. The use of silver has not diminished but among 196 nationwide Mussel Watch sites sampled in at least six years since 1986, silver concentrations showed a decreasing trend ($p<0.05$) at 24 sites and an increasing trend at 1. However, because of a change to neutron activation from a more sensitive atomic absorption method, 16 of the 24 decreasing trends had a zero (not detected) concentration at the end of the time series. Results of reanalysis of samples collected through 1994 following extraction in excess chloride (Daskalakis et. al, 1997), included those from 132 sites with 6 or more years of data. These data, not dominated by non-detects in the last year, showed decreasing trends for silver at 15 sites and 1 increase. In summary there is a case, albeit weak, for an overall decreasing trend in silver.

PWA083 Characterization of Chelating Resins for Use as Competing Ligands in Determination of Silver Speciation in River Water. Herrin, R.T.; Andren, A.W.; Armstrong, D.E.; University of Wisconsin Water Chemistry Program, Madison, WI. The chemical form of silver (Ag) in natural waters is not well known. Of particular interest is the extent and nature of Ag complexation to dissolved and colloidal ligands of unknown structure, e.g., dissolved organic matter (DOM). One operational approach for characterization of Ag species in natural waters involves titration with Ag^+ in conjunction with competing ligand equilibration (CLE/Ag^+ titration). Previous CLE/titration studies have involved the introduction of a dissolved competing ligand followed by solvent extraction, or voltammetric stripping. Practical problems with these techniques have led us to use a system in which the competing ligand is immobilized on the surface of a resin. Chelex-100, whose surface contains iminodiacetate groups, was chosen for its selectivity for Ag^+ over major cations such as calcium. Before using this system to study natural waters, we performed experiments with well-defined aqueous solutions prepared in a clean laboratory so that meaningful interpretations of results from natural systems can be made. The resin was used in the sodium form rather than the ammonium form to avoid complications caused by silver-ammonia complexes. A sodium acetate-acetic acid buffer was chosen to study pH effects, since stability constants for Ag-acetate complexes are much lower than, for example, Ag-TRIS complexes. Effective stability constants for Ag with resin-bound iminodiacetate complexes were determined by performing CLE/Ag^+ titrations on solutions containing dissolved ligands with well-defined stability constants for complexes with Ag (e.g., glutathione, penicillamine). Comparisons of experimental results to equilibrium speciation models indicate that effective stability constants of resin-bound iminodiacetate ($\text{Log } K \sim 5.3$) are approximately an order of magnitude higher than those of dissolved alkyliminodiacetates ($\text{Log } K = 4.3$).

PWA084 Fate of Silver at an Old Mining Camp, Cobalt, Ontario. Adams, N.W.H.; Collins, P; Kramer, J.R.; Manolopoulos, H., McMaster University, Hamilton, ON. Silver concentrations in water, sediment and plant material were assessed at Cobalt, Ontario. Cobalt, now inactive, produced over one-half of the World's Ag (also Co etc) 30 - 100 years ago. Clean sampling and analysis conditions with a large quality control assemblage (25% of total samples) were used to obtain reliable data. Aqueous concentrations (<10 kDa) were often <50 pico-molal (<5 ng L^{-1}). Filterable Ag occurred predominantly in the colloidal (0.2 µm - 10 kDa) size. Acid volatile sulfide persists in oxidizing waters in excess over Ag. Silver is taken up rapidly by sediment and roots of plants. Bio-accumulation of Ag(I) can be discerned only in 100 yr old cattails (*Typha latifolia*) at one location. There was no evidence of Ag(I) accumulation in market vegetables planted on the tailings. We conclude that Ag(I) occurs in ng L^{-1} levels in solution as Ag-S(II-) colloids, is rapidly and strongly and irreversibly bound to sediment and is not readily available to plant material.

PWA085 Good Ecological Risk Assessment Practices. Chrostowski, P.C.*, Foster, S.A., Durda, J.L., Preziosi, D.V. The Weinberg Group, Inc. Washington DC. Ecological risk assessment is a technique that has been in use for over a decade in regulatory decision making. The level of confidence in risk assessment results, however, can fluctuate widely depending on the audience and intended use of the analysis. This lack of confidence has often resulted in regulatory agencies making overly conservative decisions. Although some authors and regulatory agencies have developed consensus protocols for performing focused parts of ecological risk assessments, a comprehensive specification of good ecological risk assessment practice does not exist. We are proposing to fill this gap through the development of the good ecological risk assessment practices (GERAP) presented in this paper. Developed by analogy to other "good practice" approaches and in parallel to the good risk practices we have developed for human health risk assessment, the GERAP principles presented in this paper are not intended to standardize methodology, but to formalize the ecological risk assessment process. The principles encompass all critical stages in the risk assessment process starting with defining objectives and concluding with quality assurance and peer review. They also incorporate a tiered framework that allows the rigor of the assessment to be commensurate with the purpose and objectives of the analysis. Both the GERAP principles and case studies will be presented in the paper.

PWA086 The effects of pollutants on detritus processing: A case study utilising novel molecular probes. Bermingham S*, Clayton S & Maltby L. Department of Animal & Plant Sciences, University of Sheffield. Leaf litter is an important primary energy source in Northern temperate streams and rivers. Its decomposition is the result of interactions between the leaf, aquatic fungi and detritivorous invertebrates. While this process is crucial to the ecology of fresh waters there have been few studies to assess how it is affected by pollution. Studies of been limited by the lack of appropriate techniques to identify and quantify the key organisms in this process, namely the aquatic fungi. Recently novel molecular probes have been developed which can identify and quantify individual fungal species. The effect of a number of point discharges on leaf processing rates, fungal assemblages (using both conventional methodology and molecular probes), macroinvertebrate communities and detritivore feeding was investigated. Detrital processing was significantly reduced downstream of the majority of discharges studied. Detritivore feeding was positively correlated with leaf processing rates. Fungal assemblages were also affected with fewer species and lower fungal biomass downstream of the discharges. There were however some discrepancies in the results from the two methods employed to study the fungal assemblage.

PWA088 Selenium bioaccumulation and exposure in an ephemeral pool environment. Byron, E.R.*, Santolo, G.M., Ohlendorf, H.M., CH2MHILL, Sacramento, CA; Delamore, M., U.S. Bureau of Reclamation, Fresno, CA. Kesterson Reservoir, in the Central Valley of California, became famous in the early 1980s as a site of selenium toxicity to waterfowl as a result of the reservoir storage of selenium-enriched agricultural drainwater. The eventual solution to the problem required halting new sources of drainwater, followed by draining and partially filling the reservoir to convert it to terrestrial habitat (with off-site mitigation for lost wetlands habitat). The goal was the elimination of wildlife exposure to toxic levels of selenium through aquatic pathways. Selenium concentrations in winter rainwater pools that form within the terrestrial habitat have been monitored annually since 1990. The monitoring results from the seasonal pools provide a unique opportunity to estimate mobilization of selenium from soil to water and bioaccumulation of selenium from a spatially-limited, and seasonally-ephemeral source. Monitoring results include selenium concentrations in invertebrates as well as water. The seasonal and areal extent of ponding was determined from a series of aerial photographs. This poster depicts the extent and spatial pattern of recent winter ponding on the site, the concentrations of selenium in the water, and estimates of the exposure of shorebirds and waterfowl to selenium in water and from their consumption of invertebrates living in the pools. The monitoring results were combined into an index of exposure to waterbirds based on the areal extent of ponding, the selenium concentrations in the ponds, the relative abundance of the birds and their feeding behavior in various habitats, and the duration of seasonal ponding. The index provides a comparison among years of selenium exposure to the birds through the ephemeral pool pathway.

PWA089 Hickam Air Force Base: A Case Study of Stakeholder Involvement and Early Decision Processes for addressing Sediment Exposure in an Ecological Risk Assessment. Saban, L.B.*, CH2M HILL, Bellevue, WA; Townley, P.J., CH2M HILL, Bellevue, WA; Barry, W., 15th Air Base Wing, Environmental Restoration Program, Hickam AFB, Hawaii; Grannis, W., 15th Air Base Wing, Environmental Restoration Program, Hickam AFB, Hawaii. A streamlined risk assessment performed for two active stormwater drainage canals at Hickam Air Force Base, Hawaii characterized ecological risks with sufficient confidence to enable project stakeholders involved in the decision process to quickly determine appropriate responses: no further action, or further evaluation and/or presumptive remedies under the Engineering Evaluation/Cost Analysis (EE/CA) process. The EE/CA process is designed to streamline the more detailed and lengthier Remedial Investigation/Feasibility Study (RI/FS) process. This evaluation identified and focused on the issues of greatest concern and the ecological resources at greatest potential risk in order to expedite decision documents and, where necessary, preliminary cost-benefit analysis of potential remedial alternatives. The streamlined risk evaluation developed initial conceptual exposure models for the canal sites to identify assessment endpoints; and measures of exposure and effects predictive of the assessment endpoints. Risk thresholds–preliminary benchmarks to determine whether further evaluations (of causation and sources) are warranted–were then developed for these endpoints. The risk thresholds provide clear decision points to help obtain early consensus from risk managers and other project stakeholders involved in the decision process. As part of the design of the ecological risk assessment, and discussed at length during stakeholder meetings, was the use of mainland sediment bioassay organisms in Hawaiian waters. After evaluation of the ecology of the organisms used in these sediment bioassay tests, two organisms, *Neanthes arenaceodentata* and *Ampelisca abdita* were selected for use in this project. *Neanthes* had not previously been used, to our knowledge, in Hawaiian waters. The use of *Neanthes* appeared appropriate for these canals based on the ecology of these organisms and the physical characteristics of the canals. Preliminary results indicate Neanthes performs fairly well in Hawaiian waters, while Ampelisca may perform well under limited environmental conditions.

PWA090 Site-Specific Evaluation of Ecological Risks from Sediment Contamination in an Urban Wetland. C. Mach, C. Comer, and P. Geiger, Ecology & Environment; S. Wenger, Adolfson & Associates; and C. Choy, C. Prescott, and M. Abrams, Portland Bureau of Environ. Services. A site-specific evaluation was conducted for a wetland in urban Portland, OR where metal levels in sediment near several stormwater outfalls were found to exceed benchmarks for the protection of benthic life and/or wildlife. To determine the significance of the problem, additional field studies were conducted to define the nature and extent of contamination and its potential ecological effects. Sediment was collected from equally-spaced locations in the wetland and at the outfalls in the wetland and measured for chemicals of concern (metals, semivolatile organic compounds, petroleum hydrocarbons, pesticides, and PCBs), toxicity to midge larvae, and parameters that affect contaminant bioavailability in sediment. Sediment in the wetland was found to be toxic to midge larvae and survival and growth of the larvae were correlated with the sediment concentrations of several pesticides and petroleum hydrocarbons, which appear to be entering the wetland from nearby farms and newly developed areas in the drainage basin. This study shows how site-specific data can be used to better define the magnitude of an ecological problem, determine the most likely causative agents of the problem, and help prioritize pollutant source identification and control activities for a small watershed.

PWA091 Comparing Ecological Impacts From Various Remedial Actions. Friant, S.L.*, Rockel, Miller, G. ENTRIX, Inc., Wilmington, De and Bent, T.A. Bridgestone/Firestone, Nashville, TN. How many times has the question been asked "Is the cure worse than the disease, or is the risk of remediation likely to create a greater risk than no action?" Once human health risks are eliminated or managed then ecological risks can become the major clean-up driver. To decide whether a site is to be remediated based on ecological risks do require that the question be asked "Is the cure worse than the disease?" To effectively manage ecological risks the question must be addressed. This paper will present a framework and method for evaluating ecological impacts from remediation or ecological risks from remediaton. The framework is Net Environmental Benefits Analysis (NEBA) which uses habitat equivalency analysis (HEA) to quantify changes in ecological services from various remedial options. The NEBA framework uses ecological restoration science and natural resource economic methods to develop a common metric or ecological currency for comparing impacts from remedial options. This impact or environmental debit is expressed in unit of service acre years (SAY). A comparison of environmental debits between various remedial options allows for the selection of the most environmentally friendly option, yet still meeting the environmental clean-up objectives. This paper will present several examples of the framework and methods used to complete the analysis.

PWA092 The Impact of Treated Effluent from a Passive Dewatering Facility to Human Health and Aquatic Life. Bukhala, U.*, Garibay, R., The ADVENT Group Inc., Rosslyn, Virginia; Volanski, J., US Steel, Pittsburgh, Pennsylvania. During the proposed USS dredging of the East Branch of the Grand Calumet River, the contaminated sediments will be secured in a Passive Dewatering Facility (PDF). The treated PDF effluent will be returned to the river. The projected chemical characteristics of the treated effluent has the potential to cause a temporary exceedance of the facility's water-quality based effluent limits (WQBELs) for some parameters. In response, a variance from the water quality standards has been requested. As a part of the variance application process, the Indiana rules require the discharger to assess the impact of the parameters to Aquatic Life and Human Health. The impact is assessed by comparing the projected downstream instream concentrations to the applicable criteria. If the projected downstream concentration exceeds the criterion, then a risk analysis is performed: For the aquatic life impact, a comparison of the predicted downstream instream concentrations to aquatic thresholds for resident species is performed; and, For the human health impact, the cancer risk posed by the treated effluent is evaluated. It was determined by this study that most of the parameters with WQBELs based on Aquatic Life Criteria, will be discharged below aquatic life threshold values and the parameters evaluated for cancer risk will be below the acceptable Indiana risk factor.

PWA093 **Geographic Information System (GIS) Evaluation and Analysis of Sediment Risk Assessment Data in Pearl Harbor, Hawaii.** Lester, W.C.*, Clayton, J.R., Jr., and Blackwelder, R.B., Ogden Environmental, San Diego, CA; Nakamura, P., Naval Facilities Engineering Command, Pacific Division, Pearl Harbor, HI; Grovhoug, J.G., SPAWAR Systems Center, San Diego, CA. Risk assessments for chemicals in marine sediments in Pearl Harbor, Hawaii require evaluation and analysis for the spatial nature and extent of chemicals of concern in sediments, the occurrence and location of habitats for biological receptors of concern, and the completeness of exposure pathways for chemicals from sediments to receptors. Application of GIS procedures to risk assessments in an interactive mode can greatly enhance the overall process for initial conception/design, evaluation of risk estimates, and clean-up alternatives. Initially, GIS is used to develop the sampling design. GIS supports identification of discrete sampling areas that encompass considerations for bathymetric, physiographic, and sedimentological characteristics, present and historic inputs of chemicals from associated watersheds, known or suspected point sources of chemicals, and habitats of biological receptors. GIS is used to select random sampling locations within sampling areas and generate coordinates of sample sites to facilitate field sampling. During analysis, GIS is used to display and quantify spatial extents of chemical distributions and risk values that incorporate chemical concentrations for sediments, characteristics of the natural history of specific receptors, and the spatial extent of sampling areas. Risk values and their distributions, which are the desired result for the risk assessment process, are not necessarily the same as chemical distributions. Finally, the spatial extent and distribution of risk for individual areas of sediment is used to evaluate and guide remediation considerations, which provides a quantitative estimate of the overall improvement in environmental quality associated with each clean-up alternative.

PWA094 **Development of Measured Site-Specific Biota-To-Sediment-Accumulation-Factors (BSAFs) to Support Risk Assessments of Marine Sediments in Pearl Harbor, HI.** Clayton, J.R., Jr.* and Lester, W.C., Ogden Environmental, San Diego, CA; Nakamura, P., Naval Facilities Engineering Command, Pacific Division, Pearl Harbor, HI; Grovhoug, J.G., SPAWAR Systems Center, San Diego, CA. Measurement-based site-specific BSAFs are developed for five types of sediment-associated aquatic receptors in Pearl Harbor (infauna, crabs, tilapia, goatfish, and gobies) based on chemical measurements in paired samples of whole wild-caught organisms and sediments. BSAFs are developed for 145 chemicals including metals, organotins, polynuclear aromatic hydrocarbons, chlorinated pesticides, PCBs, and polychlorinated dibenzo-p-dioxins/furans. The samples of wild-caught organisms were collected from 15 locations and the sediment samples from 97 locations in the harbor. BSAFs for relatively stationary organisms (infauna, crabs, and gobies) are calculated as concentration ratios in tissue samples to those in sediments from the proximity of the tissue collections. BSAFs for mobile organisms (tilapia and goatfish) are calculated as concentration ratios in tissue samples to spatially weighted concentrations of sediments that incorporate estimates of habitat usage areas for the particular organism. BSAFs are calculated on the basis of two normalization approaches: dry-weight-to-dry-weight chemical concentrations in tissue and sediment samples and concentrations normalized to lipid content for tissues and total organic carbon for sediments. Variability in BSAFs is considered for the two normalization procedures. The actual measured tissue concentrations are compared to tissue values estimated from literature-derived equilibrium partitioning constants (sediment-to-water K_{oc} and water-to-tissue BCF values) applied to the measured sediment concentrations. The BSAFs are being used in ecological and human health risk assessments for sediments in Pearl Harbor. Use of the BSAFs provide site-specific estimates for sediment-related exposures to chemicals for target biological receptors.

PWA095 **Integration of Chemistry, Toxicity, and Bioaccumulation Estimates for an Effects-Based Ecological Risk Assessment of Marine Sediments in Pearl Harbor, Hawaii.** Clayton, J.R., Jr.* and Lester, W.C., Ogden Environmental, San Diego, CA; Nakamura, P., Naval Facilities Engineering Command, Pacific Division, Pearl Harbor, HI; Grovhoug, J.G., SPAWAR Systems Center, San Diego, CA. Chemicals in marine sediments in Pearl Harbor, Hawaii may pose risk to ecological receptors. Assessing risk requires field measurement of the nature and extent of chemicals in sediments, biological receptors at risk, and the completeness of exposure pathways between sediments and receptors. Risk estimates are generated from receptor-specific information for chemical-specific estimates of exposure, effects, and risk. Evaluation is given to both direct toxicity for sediment chemicals and bioaccumulation to higher trophic levels. Direct toxicity is assessed by exposing test organisms to sediments in laboratory bioassays. Statistically significant increases in toxicity with increasing chemical concentrations are used to demonstrate toxicity-chemical relationships. Risk for bioaccumulation requires understanding of site-specific foodweb relationships. Estimates for mobile receptors incorporate information for relative contributions of sediments from different locations. Development of measurement-based site-specific biota-to-sediment-accumulation-factors for chemicals in aquatic receptors is used to increase the accuracy of exposure estimates for aquatic and higher trophic level receptors. Risk is quantified as a chemical-specific toxicity quotient, which is the ratio of a measured sediment-based exposure point value to an appropriate literature-derived toxicity reference value. Variability for risk calculations is addressed by Monte Carlo methods to develop probabilistic estimates of risk. The major advantage in applying effects-based risk techniques is generation of realistic, site-specific estimates for risk based on considerations for direct toxicity of sediments, bioaccumulation of chemicals to higher trophic levels, and habitat usage areas by receptors of concern.

PWA096 **Multipathway Human Health Risk Assessment for Marine Sediments in Pearl Harbor, Hawaii.** Thompson, V.L.*, Mahini, X. P., Lester, W. C., and Clayton, J. R., Jr., Ogden Environmental, San Diego, CA; Nakamura, P., Naval Facilities Engineering Command, Pacific Division, Pearl Harbor, HI. Chemicals detected in marine sediments in Pearl Harbor, Hawaii may pose risk to human receptors through a variety of exposure pathways. The first step in assessing potential human health risks requires an evaluation of the nature and extent of chemicals in marine sediments and edible marine organisms such as crabs and fish. Chemical concentrations in sediments were measured in samples collected from Pearl Harbor. Chemical concentrations in edible species of crabs and fish were estimated using both field sampling data and a bioaccumulation model to estimate the effects of exposure of crabs and fish in Pearl Harbor to chemicals in marine sediments. The second step in assessing potential human health risks requires estimating potential exposure to humans through direct contact with chemicals in marine sediments and exposure through ingestion of edible crabs and fish. Data on ingestion of fish by Hawaii residents were obtained from a thyroid cancer study conducted by the University of Hawaii Manoa. These data were used to estimate ingestion rates by humans of Pearl Harbor crabs and fish in exposure scenarios ranging from occasional sport fishing to subsistence fishing. Uncertainties in risk calculations were evaluated using Monte Carlo methods to develop probabilistic estimates of risk. Human health risk results were integrated into a harbor-wide evaluation of chemicals in marine sediments using Geographic Information System (GIS) procedures. GIS combines information on the spatial extent of sediment chemicals, potential exposure of edible marine organisms to sediment chemicals, and use of the area by humans. GIS provides a display of risk estimates and their reduction for selective clean-up alternatives.

PWA097 **Ecological Risk Assessment in a Voluntary Action Program,** J. Bleiler, ENSR, Acton, MA, and D. Scott, C. Dickerson. A chemical manufacturing facility recently completed a phased closure of a facility located near a tidal river in southeastern New England. The property owner agreed to conduct a voluntary RCRA facility investigation. The immediate objective of this effort is to achieve site stabilization. Ultimately, the objective is to achieve closure under RCRA. The ecological risk assessment was designed to evaluate potential threats to the environment in two phases: on-site and off-site. Sediment chemistry, toxicity tests, and tissue analyses data were evaluated in the initial on-site phase of ecological risk assessment. In order to integrate ecological risk results with development of risk management alternatives, a range of preliminary remedial goals and alternatives were developed for several stressors, including lead, PCBs, mercury, and chlorinated benzenes. Portions of the site were designated as warranting remedial actions. The selected remedial alternatives reflected the measurement and assessment endpoints considered in the risk assessment. The second phase of work focuses on off-site transport of bioaccumulative compounds of concern. Sediment analyses include detailed nature and extent evaluations, including PCB and mercury investigations, and sediment core sampling. Elevated levels of mercury were found in the offsite coastal embayment. Biological sampling efforts for the offsite effort are currently being planned.

Because the risk assessment at this site is being conducted under a voluntary action program, the property owner has been able to effectively manage the program schedule and scope. This voluntary risk assessment will be used as a key tool to help provide a means for reaching consensus between risk assessors and risk managers. The presentation will discuss the pros and cons of the voluntary action program experience at this site.

PWA098 Use of Halophytes for the Cost-Effective Assessment and Remediation of Oilfield Brine Impacts in Arid Terrestrial Ecosystems. P. Rury*, D. Turton, Arthur D. Little, Inc., Cambridge, Massachusetts, and D. Owens, Dow, Cogburn and Friedman, Houston, Texas. Electrical conductivity of non-saline soils, lower than 2 deciSiemens per meter (dS/m), is tolerated by salt-sensitive crops. Halophytes are plant species that tolerate soil salinity greater than 8 dS/m and crops that yield satisfactorily above this level. Soil salinity impacts, floristic changes, and terrestrial ecological damages from produced water releases are a function of precipitation, hydrology, soil quality, and salt tolerance of plant communities at oilfield sites. Natural recolonization of brine-impacted soils by halophytes (e.g., family Chenopodiaceae) is most critical in arid habitats where precipitation cannot leach soil salts to below the rooting zone. Halophyte recognition helps focus parsimonious soil sampling to yield desired salinity impact data without the high costs of grid-based sampling programs otherwise needed when visible evidence of produced water releases (e.g., salt or petroleum stains) is lacking. Litigation-related mapping of brine-impacted soils and rangeland vegetation at a West Texas oilfield confirmed that halophytes are reliable indicators of saline soils. Soils were collected and floristic data recorded from potential brine impact areas which either lacked vegetation or supported combinations of halophytes and/or non-halophytes, including range grasses cultivated for grazing cattle. Soils with average salinity of 38 dS/m (maximum of 82 dS/m) were recolonized only by halophytes (e.g. *Suaeda*), whereas non-halophytes (e.g., *Eragrostis Lehmanniana*) recolonized soils with average salinity of 3.4 dS/m (maximum of 11.3 dS/m). Future colonization by halophytes is expected on bare soils with maximum salinity of 76 dS/m. These results confirm the reliability of halophytes for focusing soil sampling efforts to assess oilfield brine impacts and their ecological restoration value for use in saline soil phytoremediation.

PWA099 Use of Food Chain Models and Ecological Risk Reduction Curves to Develop and Apply Wetland Sediment Cleanup Goals. P.M. Rury*, D.J. Turton, Arthur D. Little, Inc., Cambridge, Massachusetts, S.C. Svirsky, U.S. Environmental Protection Agency, Boston, Massachusetts, and K. Munney, U.S. Fish and Wildlife Service, Concord, New Hampshire. An ecological risk assessment (ERA) was performed to guide remedial design for polychlorinated biphenyls (PCBs), pesticides, chromium, and lead in a red maple swamp at a New England Superfund site. A 100 foot grid was sampled for PCBs and a geographic information system (GIS) was used to contour PCB concentrations (maximum of 300 mg/kg) and calculate a spatially-weighted average Total PCB concentration for each contoured segment and for the entire wetland (weighted mean of 1.4 mg/kg). Deterministic and probablistic (Monte Carlo) food chain exposure models, pre-calibrated with site-specific bioaccumulation factors for earthworms and fish, were used to realistically estimate exposures and develop cleanup goals for the short-tailed shrew, American woodcock, and mink. After the Monte Carlo analysis identified the most likely foraging scenarios for the wide-ranging mink and woodcock, deterministic models were used to calculate sediment cleanup goals for all contaminants and species. Given the logistical difficulties of forested wetland remediation, the most realistic exposure models were then used to calculate residual risks from contamination that might remain after focused remediation.These models were used to prepare species-specific, PCB risk reduction curves for alternative, Total PCB cleanup goals of 1 mg/kg and 10 mg/kg. With sediment remediation focused on the most accessible 4.8 acres with weighted mean Total PCBs exceeding 10 mg/kg and assuming a residual Total PCB sediment concentration of 1 mg/kg, 70% of the Total PCB sediment risk can be eliminated, resulting in a residual Total PCB hazard quotient of less than 1.5 throughout the 60 acre wetland, for all three species.

PWA100 Risk Contours Used to Delineate Safe/Unsafe Chemical Aerosol Concentrations for *Myotis sodalis* and *Myotis grisescens*. Schmidt, A.*; Salyers, J.; and Romme, R. 3D/International, Inc., Environmental Group; Cincinnati, Ohio. Certain military installations use aerosols that may be toxic to endangered species. Military aerosols are commonly released from grenades, smoke pots, and generators and function as obscurants for troops and equipment. To determine the toxicity of aerosols and at what concentrations they cause adverse toxicological effects, toxicity threshold values were determined for specific aerosols being studied. Threshold values are concentrations that are expected to result in adverse toxicological effects. We selected NOAEL (No Observable Adverse Effect Level) as our toxicological benchmark value. Specific *Myotis sodalis* and *Myotis grisescens* NOAEL values were not available for any of the aerosols under investigation. NOAEL values for *Myotis sodalis* and *Myotis grisescens* were estimated by normalizing (adjusting for differences in doses based on body weight) values reported for standard laboratory test animals (e.g., rats, mice, guinea pigs). The normalized NOAEL values for the two species of bat were used in an air dispersion model to determine the distance the NOAEL concentration travels from release points. Down wind and crosswind dispersion were modeled. Risk contours were developed to depict areas where toxicity threshold values may be exceeded. Chemical release guidelines were based on developed risk contours. These contours indicate how far away from a hibernacula or maternity colony an aerosol release could occur without causing toxicological effects to bats.

PWA101 Ecological Risk Assessment of Lead in a Reservoir Subject to Water Level Fluctuations. Mitchell, D.F.*, Wandland, K.D., ENSR Consulting, Acton, MA and Wade, D.C., Tennessee Valley Authority, Mussel Shoals, AL. An ecological screening risk assessment (ESRA) was conducted by the Tennessee Valley Authority (TVA) to evaluate potential adverse effects of lead on ecological receptors and resources at or near the Appalachian Smelting and Refining Company (ASRC) Site at the South Holston Reservoir, Sullivan County, Tennessee and to evaluate potential remedial options at the site. The purpose of the ESRA was to review and evaluate data and test results for the ASRC Sites to determine if lead-contaminated media (i.e., slag road materials, battery remnants) at the former battery reclamation site posed a potential adverse risk to aquatic receptors (fish, benthos, wildlife, habitats) on the shoreline or within the confines of the reservoir. Due to seasonal inundation of the site (i.e., annual drawdown can exceed 50 feet per year), no terrestrial receptors were at risk. Risk characterization indicated no risk posed to aquatic organisms in the water column. Weight-of-evidence evaluation of multiple measurement endpoints for sediment (i.e., bulk concentrations, porewater, SEM:AVS, and toxicity tests) demonstrate little overall adverse risk associated with sediment exposure. Finally, food web modeling of higher trophic avian (kingfisher, great blue heron, bald eagle) suggested limited potential concern for avian receptors at one of the two areas of concern. The overall results of the ASRC ESRA, interpreted by a weight-of-evidence evaluation, indicated no potential ecological concern for the aquatic receptors and low potential concerns for semi-aquatic avian receptors at Site 1. Planned remediation through removal of slag material at Site 1 would eliminate the low potential ecological concerns there.

PWA102 Ecological Risk Analysis for Sandhill Cranes Staging on the Platte River, NE. Parkhurst, B.R.*; Kennedy, J. The Cadmus Group, Inc., Laramie, WY. The U.S. EPA is conducting ecological risk assessment case studies to help develop guidance for applying ecological risk assessments to both chemical and non-chemical stressors on a watershed scale. One of these case studies examines the Middle Platte River, NE. The assessment endpoint was Sandhill Crane abundance and distribution. The Middle Platte River floodplain plays a key role in the life cycle of Sandhill cranes. Approximately 500,000 cranes spend upwards of six weeks staging on the middle Platte River floodplain and adjacent agricultural land. Sandhill cranes roost in open river channels and forage for invertebrates in nearby wet meadows and for waste corn in cornfields. The cranes potentially are at risk from loss of habitat, in particular, roost sites and wet meadows, caused by decreased river flows. The goal of this project was to develop a model to predict changes in Sandhill crane abundance and distribution as a function of the quantity and distribution of habitat. To develop such a model, we analyzed data on crane habitat and abundance from 1978 to 1996 along the Platte River floodplain. The data were evaluated at two geographical scales, a smaller one based on bridge segments, and a larger one based on delineated staging areas. On the bridge segment scale, relationships between the area of key habitat types, such as wet meadows and optimum roost sites, and Sandhill crane densities were weak. On the staging area scale, significant relationships were found between Sandhill crane densities and mean channel width, the density of wet meadows, and the density of alfalfa fields. Based on this information, a model was developed that estimates crane density as a function of these habitat parameters.

PWA103 Risk-based Prioritization of a Former Refinery. Peterson, S.C.*, Comer, C.E., Freeman, R.W., Stineman, C., Castle, D., Brown, N., Ecology & Environment, Lancaster, NY; Jackson, L.J., Beatty, P., Lyverse, M.A., Chevron Research & Technology Company, Richmond, CA; Tiffany, J.C., Chevron Products Company, Hooven, OH; Kearns, D.B., Boehnker, D.W., QST Environmental, Inc., Miamisburg, OH. A risk-based approach was developed to prioritize remedial decisions at a former Chevron petroleum refinery in the vicinity of Cincinnati, Ohio. The site occupies several hundred acres adjacent to the Great Miami River and is located near residential communities. Ecologically sensitive areas on the perimeter of the former refinery site include mature bottomland forest and emergent wetlands. The site is undergoing a RCRA Facility Investigation (RFI) to address a diesel and leaded gasoline plume on the groundwater; refinery waste disposed at various locations; and other issues related to petroleum hydrocarbons and metals. Innovative methods of site characterization included the use of Laser-Induced-Fluorescence (LIF)for subsurface soil delineation, and select ion mode (SIM) GC/MS. Multi-tiered ecological and human health risk assessments are being performed to evaluate the potential impacts of offsite and onsite contaminant migration and exposure. A land use plan was developed to provide a framework for evaluation of future use scenarios appropriate for the facility. Available data were evaluated to identify areas of the site that pose the greatest potential risk. Additional soil and groundwater data were collected from these areas and risks were characterized based on a site-specific screening approach. The approach was directed at identifying areas that might pose an unacceptable risk for realistic future uses, rather than the more typical approach of "screening out" areas of concern using generic assumptions and levels of *de minimis* risk. The prioritization was used to identify the need for interim measures and to help focus further investigations.

PWA105 Design of an Ecological Risk Assessment for a Former Naval Shipyard with Seasonal Wetlands and Dredge Disposal Areas. Dubinsky, E., TtEMI, San Francisco, CA; Bernhard, T.S., EFA West, U.S. Navy, San Bruno, CA; Baker, W., EFA West, U.S. Navy, San Bruno, CA; Linder, G., TtEMI, San Francisco, CA; Gleason, M., TtEMI, San Francisco, CA; Brown, R.W., TtEMI, San Francisco, CA; Demetrios, V., TtEMI, San Francisco, CA. Mare Island is a decommissioned naval shipyard in the northern San Francisco Bay that contains upland habitat and natural and created wetlands that are used by a variety of ecological receptors. Preliminary site assessment results indicated the possibility of multiple environmental stressors, including both chemical and physical factors. Habitat surveys identified fresh water and brackish wetlands, and coastal salt marsh. Most of this site is comprised of fill material and large areas have been used for the disposal of dredge materials. Some dredge spoil ponds, which are enclosed by berms that preclude tidal influence to the area, contain wetland vegetation typically found in tidal marsh areas. Other dredge ponds on this site are actively managed to preclude wetland vegetation from becoming established. The unique nature of many of these habitats and the variation in salinity between wetland areas and seasonally inundated dredge spoil ponds called for the use of a variety of measurement endpoints. Soil and sediment bioassays and marine and freshwater aquatic bioassays were conducted to evaluate potential toxicity of various media at the site. Soil, sediment, and surface water chemistry samples were collected along with collocated tissue samples of plants, aquatic invertebrates, amphibians, and small mammals to evaluate bioaccumulation and food chain effects.

PWA106 Bisphenol A: A Review of the Environmental Fate, Effects, and Exposures. Dorn, P. B.,* Shell Chemical Company, Houston, TX; Staples, C. A., Assessment Technologies, Inc., Fairfax, VA; Klecka, G. M., The Dow Chemical Company, Midland, MI; O'Block, S. T., Aristech Chemical Corporation, Pittsburgh, PA; Branson, D. R., GE Plastics, Mt. Vernon, IN; Harris, L. R., Society of the Plastics Industry, Washington, DC. Bisphenol A (CAS 80-05-7) is used in the production of polycarbonate and epoxy resins, in adhesives, coatings and other applications. During manufacturing 0.017% of BPA production may be released as fugitive air emissions, to surface waters, to wastewater treatment plants, 0.085% of the release is recycled, landfilled or incinerated. BPA is moderately water soluble (120,000 to 300,000 $\mu g/L$ at pH 7), may adsorb to sediment (K_{oc} 314 to 1524), has low volatility, but is rapidly biodegraded in both acclimated wastewater treatment plants and receiving waters (half-lives 2.5 to 4 days) as well as the OECD ready biodegradability test. A Mackay Level I model estimates BPA environmental distributions to be 52% to water, 23% to sediment, 25% to soil and de minimus amounts (<1%) in other compartments. BPA is toxic (lowest algal EC50 of 1000 $\mu g/L$) but has low potential for bioaccumulation in aquatic organisms (BCFs 5 to 68). The chronic NOEC for *Daphnia magna* is >3146 $\mu g/L$. Surface water concentrations from the literature and recently collected around the US manufacturing locations are one to several orders of magnitude lower than concentrations causing chronic effects, with most concentrations nondetected (< 1µg/L). The PEC/PNEC ratios show a 3 to >80 margin of safety. The assessment of available literature and recently collected data on exposures in receiving waters and effects suggests BPA presents minimal ecological risk. Further studies are in progress to reduce uncertainty in this assessment.

PWA107 An Environmental Risk Assessment of Bisphenol A in U.S. Surface Waters. C.A. Staples*, Assessment Technologies, Inc. Fairfax, VA, Philip B. Dorn, Shell Development Company, Houston, TX, Dean R. Branson, GE Plastics, Mt. Vernon, IN, Gary M. Klecka, The Dow Chemical Company, Midland, MI, Sondra T. O'Block, Aristech Chemical Corporation, Pittsburgh, PA, and Jennifer Hayward, Society of the Plastics Industry, Washington, DC. Bisphenol A is used in manufacturing epoxy and polycarbonate resins (640 million kg, 1995 U.S. production). About 2500 kg of bisphenol A (BPA) were reported released from U.S. producers and processors in 1995 to surface waters following treatment. For product stewardship reasons, BPA surface water concentrations were measured upstream and downstream of producers and processors of BPA. BPA is readily biodegradable (81 to 93% of the theoretical oxygen demand in 28 days) and river die-away half-lives range from 2.5 to 4 days. An analysis of estimated surface water discharges with well known stream flows (7Q10) corresponds to significant margins of safety based on chronic no effect concentrations. An analytical method was developed that was capable of detecting 1 µg/L BPA (>1000-fold lower than all measured chronic no effect concentrations) and that did not produce false positives by inadvertent conversion of BPA adducts to BPA. The method utilized cool on-column injection with GC/MS. Concentrations of BPA were measured upstream and downstream of seven manufacturing and processing facilities in the U.S. Samples were collected in the late summer to fall of 1996 and 1997, approximately the low receiving stream flow periods. Nearly all surface water samples upstream and downstream of each facility showed no detectable levels of bisphenol A, based on a detection limit of 1.0 µg/L. These results are finally compared with other monitoring data and are evaluated in the context of an environmental risk assessment.

PWA108 Application of the Screening Level Ecological Risk Assessment Protocol for a Hazardous Waste Burning Facility. Yurk, J.J.*, U.S. EPA, Dallas, TX; Desmond, W.P., Tetra Tech EMI, Dallas, TX; Matthews, B.M., Tetra Tech EMI, Dallas, TX. Operating permits for hazardous waste burning combustion facilities are required under the Resource Conservation and Recovery Act (RCRA) to be protective of human health and the environment. A *Screening Level Ecological Risk Assessment Protocol for Hazardous Waste Burning Combustion Facilities* (SLERAP) was developed by U.S. EPA, 1998 as a tool to be used to estimate potential risk to the environment posed by stack and fugitive emissions generated during current and proposed future operation of a hazardous waste burning facility. The screening ecological risk assessment results provide regulatory permit writers with information by which they can assess writing permit conditions protective of the environment. A generic ecological risk assessment was conducted to illustrate the use of the SLERAP. The case study walks the reader through the combustion ecological risk assessment process including: (1) selection of chemicals of concern; (2)

defining the habitats to be evaluated; (3) selecting the habitats most vulnerable to exposure to combustion emissions; (4) food web construction; (5) defining critical attributes for protection of the environment, commonly referred to as assessment endpoints; (6) measurable endpoints, commonly referred to as measures of effect; and (7) permit applications of results. Permitting options/conditions based on results of a screening ecological risk assessment can include; limits on facility operating conditions, waste feed limits, environmental monitoring requirements, and/or denial of a permit.

PWA109 Comparative Physical Risk Analysis for Cost-Effective Environmental Management: The Teller Fencing Case Study. Mahini, X. P., Ogden Environmental and Energy Services Co., Inc., San Francisco, CA and Vagt, W., Alaska Army National Guard, Fort Richardson, AK. The Alaska Army National Guard (AKARNG) received a Notice of Violation (NOV) from the United States Environmental Protection Agency (USEPA) for lack of security for one fuel oil aboveground storage tank (AST) at the village of Teller, Alaska. USEPA interpretation of AST regulations [40 CFR 112.7(e)(9)(i)] was that AKARNG was required to build a fence around the tank. Concerns regarding the safety aspects of fencing a site that is only manned two days a month led AKARNG to initiate a comparative physical risk analysis for the installation of a fence. Five years of state-wide accident reports were reviewed, and the risk analysis results showed fences to be more hazardous than unfenced tanks. Based on this comparative physical risk analysis, AKARNG proposed installation of motion sensor lights to comply with AST security requirements for their tanks. This innovative risk-based solution to regulatory compliance is estimated to result in savings of $41,385 dollars per year at Teller and more than $4 million per year for over 100 tanks owned by AKARNG in Alaska.

PWA110 Non-Target Organism Risk Assessment for *Bacillus thuringiensis* in Genetically Modified Plants. McKee, M.J.*: Reed, A.J., Sanders, P.R., and Nickson, T.E., Monsanto, St. Louis, MO; Sanders, P., Monsanto, St. Louis, MO. Emphasis on sustainability and the need to feed a rapidly expanding human population has presented challenges to traditional agricultural practices and technologies. Recent advances in biotechnology have allowed the introduction of desired agronomic traits through genetically modified plants. Ecological risk assessment is an important tool for evaluating these new products and can be broadly separated into two categories: 1) assessment for effects of the genetically modified plant on plant communities; and 2) assessment for potential effects on non-target organisms. The focus of this presentation is on the ecological risk assessment strategy for non-target organisms. The ecological framework used includes Problem Formulation, Risk Analysis, Risk Characterization, and Risk Management phases. The assessment strategy is demonstrated using non-target organism testing data for the *Bacillus thuringiensis* protein expressed in insect protected corn and cotton. Important aspects of Problem Formulation include screening activity towards target pests, guild analysis for potentially exposed organisms in the field, and selection of test organisms as surrogates for evaluation. The Risk Analysis phase includes an exposure assessment followed by biological effects testing on surrogates of potentially exposed animals. For insect protected cotton and corn, effects data were collected for soil organisms (i.e. earthworms and collembola); beneficial insects (i.e. honey bee larvae and adult, green lacewing, ladybird beetle, and parasitic wasps); aquatic organisms (i.e. daphnia); and birds (i.e. bobwhite quail). Risk Characterization was accomplished by comparing effects and exposure data using the risk quotient method with application of safety factors. Risk Management in this case was not necessary as no hazard was identified with the laboratory toxicity evaluation. The results of this particular protein testing program will be discussed within the context of an overall strategy for non-target organism risk assessment for genetically modified plants.

PWA111 Environmental Risk Assessment of the Priority Substance *N*-nitrosodimethylamine (NDMA) under the *Canadian Environmental Protection Act* (CEPA). Windle, W.M., Environment Canada, Ottawa, Ontario CANADA. In Canada, substances on the Priority Substances List (PSL) are assessed by two federal government departments, Environment Canada and Health Canada, to determine if they pose a risk to the environment and to human health. This presentation will discuss the ecological risk assessment only. On December 16, 1995, 25 substances were added to the PSL, including *N*-nitrosodimethylamine (NDMA). NDMA is not commercially produced in Canada nor are there any identified industrial uses. It may form naturally in aqueous systems, in soil and in nighttime air, if the precursor substances and biochemical processes are present. NDMA can also be formed as a by-product in industrial and municipal effluents when amines and nitrites are present. There are extensive acute effects data and adequate chronic effects data on NDMA. The mink, *Mustela vison*, appears to be the most sensitive wild mammalian species, whereas, the green alga, *Selenastrum capricornutum*, is most sensitive aquatic organism. A detailed risk assessment is underway to determine the extent of exposure, and the associated risks, for the Canadian environment.

PWA112 Risk Communication During the Risk Assessment Process: An Ontario Case Study. Hull, R.N.*, McKee, P.M., Fahey, J.J., Beak International Incorporated, Brampton, ON; Benson, D.C., Baker & McKenzie, Toronto, ON. Interested stakeholders (e.g., the public) are often ignored until the risk assessment (and accompanying risk management/remediation plans) are near completion. Although several regulatory jurisdictions have policies encouraging public consultation and regular communication with regulators during the risk assessment process, this activity is largely ignored or minimal effort is expended. The Ontario Ministry of Environment and Energy (MOEE) 1997 Guidelines for Use at Contaminated Sites recommends a public communication program, but does not allow regular communication between the proponent and the MOEE until final submission of the risk assessment report. Recent experience has shown that frequent communication with both the public and the regulators will greatly increase the probability that the results, conclusions and recommendations of the risk assessment will be accepted. This was illustrated during a complex human health and ecological risk assessment for a former industrial site in Ontario. Many potentially controversial issues were avoided, most likely due to regular communication. This study supports the MOEE recommendation for public communication programs. However, it also reveals that regular communication with the regulators, which contradicts the current MOEE guidance, would be beneficial not only to the proponent, but also the MOEE.

PWA113 A Web Interface to Searching the National Library of Medicine's TOXNET System. Wexler, P. National Library of Medicine; Bethesda, MD. For over 30 years, the National Library of Medicine's (NLM) Toxicology and Environmental Health Information Program (TEHIP) has been engaged in the collection and dissemination of computerized information related to hazardous chemicals, toxicology, and the environment. Among TEHIP's many files relevant to environmental toxicologists is the widely used Hazardous Substances Data Bank (HSDB), containing extensive peer-reviewed information related to ecotoxicity, pollution sources, environmental transformations and transport, human environmental exposure and exposure standards and regulations. Other important files include the EPA's Integrated Risk Information System (IRIS) and Toxic Chemical Release Inventory (TRI) series of files. These and most of TEHIP's other databases reside on the TOXNET system and have heretofore required familiarity with a specialized search language in order to successfully retrieve information. A free Web interface has recently been built to allow untrained users to easily navigate through the formidable array of data available on these files. This interface's Simple Search template permits users to specify a database, chemical name, subject term(s), and display format. Links to related sites and online help are also available. Advanced Search capabilities are being designed as are interfaces to other TEHIP files outside the TOXNET system, such as the bibliographic TOXLINE database which, with its companion file of earlier literature, contain over 2.5 million citations covering the biochemical, physiological, pharmacological, and toxicological effects of chemicals.

PWA114 Exploring Environmental Issues: Focus on Risk. Iozzi, L.*, Rutgers University, NewBrunswick, NJ; Gallagher, F., Division of Parks and Forests, Trenton, NJ; Messinger, A., Project Learning Tree, Washington, DC; Soyka, S., Project Learning Tree, Washington, DC. Participants will learn about a new secondary level curriculum:

"Exploring Environmental Issues: Focus on Risk" and how they can get involved in local educator workshops. This curriculum can be used by formal and nonmoral educators to introduce students at the high school or college level to the field of risk. Goals of the curriculum include the following: understanding how risk is defined by risk assessors; understanding how risk is evaluated, communicated, and perceived by experts and lay people; understanding that risk is a part of everyday life; identifying risks, costs, and benefits associated with environmental issues investigating potential risk issues concerning chlorine, plastics, radon, and electromagnetic fields; understanding the basics of risk assessment; applying critical thinking skills to real-world choices and policies; and recognizing the important role that risk plays in developing environmental policy, and personal risk decisions. The activities provide students with a framework through which they can apply scientific processes and higher order thinking skills to environmental issues. Many of the activity objectives meet national science content standards. Teaching students about the nature of risks, risk assessments, and risk management will help create an awareness and an understanding of the risks they face in their own environment. Once students learn the basics of risk, they should be able to apply their knowledge and skills to environmental issues and problems, public policy issues, and personal decisions.

PWA115 Data Quality for Ecological Risk Assessment. N Luke, and M. Watt, International Technology Corporation, Somerset, NJ. Ecological risk assessment has received increased emphasis in guiding remediation in Remedial Investigation (RI)/Feasibility Studies (FS). Decisions made by a risk management for an RI/FS are based on environmental data analyzed from samples collected at a contaminated site. Thus, the quality of analytical data is most critical and crucial for a remedial investigation, used for determining risks and establishing cleanup levels. In many cases, data provided to risk assessors for an RI/FS are of poor quality, resulting in expensive re-sampling, or costly delays to remedial efforts, or worse yet, unnecessary remedial actions. Up-front involvement of ecotoxicologists in sampling plan is an necessary prerequisite and an effective data quality program is the key for developing high quality of data. A data quality program needs to be implemented in all stages of an RI. These include field sampling, analytical oversight, data evaluation/validation, database development/management, and data usability and interpretation for risk assessment and other end users. Ecotoxicologists need to get involved in these various stages of a data quality program.
A case study demonstrates that an effective data quality management program and ecotoxicologists' input in ensuring collection of the data for ecological risk assessment not only resulted in a cost saving of more than $150,000, but also resolved a potential remediation dilemma.

PWA116 Hazard Assessments in the Developing World. Hinckley, D.A.*, J.M. Boltz, EA Engineering, Science and Technology, Sparks MD USA. The methodology of the assessment of risk has, to a large degree, been established in well developed 1st world locations such as the United States and Europe. Recently, risk and hazard assessment has been applied to the developing world. While in the developed world assessment of human cancer risk to 1×10^{-6} and the protection of mice and shrews on small hazardous waste site are common, such risks are trivial compared to the disease and societal implications of common practices in many developing countries. An environmental assessment is required by U.S. AID regulations for major projects, and one was produced from 1995 to 1997 for a major waste water treatment plant upgrade in the city of Alexandria, Egypt. A year long baseline characterization was completed, and used to select an alternative of choice for the upgrade. Presently waste water from Alexandria is primary treated and discharged without disinfection to a 25 km^2 lake inland of the city. While intended as a temporary solution to the city's waste water problem, this situation has been occurring since 1993, and will continue for the forseeable future. Fecal coliform concentrations in the lake ranged from from 1×10^2 to 6×10^{10}, and were commonly at 10^4 MPN/100 mL, well above any standards for Egypt, not just the developed world. Nutrient concentrations in this hypereutrophic lake averaged 2 - 3 mg/L for total P, and 10 - 12 mg/L for ammonia. The lake is largely anoxic, and no longer supports a viable fishery. The problems associated with conducting this type of baseline assessment will be discussed, and the environmental assessment conclusions placed into the context of risk acceptability and the reality of the developing world.

PWA117 Ecological Risk Assessment for Addressing Litigation Issues. Sheehan, P.*, Beach, J. and Dodge, D., ChemRisk Service, McLaren/Hart, Inc., Alameda, CA. The number of toxic tort lawsuits addressing claims of the ecological effects of chemicals has increased dramatically in the last two years. This occurrence has led to an associated increase in the opportunities provided to ecological risk assessors to evaluate ecological risks of historic, current or future anticipated exposures. In general, the focus of litigation risk assessments has been more narrow than regulatory applications. The chemicals, species and exposure pathways of interest and the relevant time period to be considered are often defined and limited by the specific claims of the lawsuit. Two examples are discussed: 1) the risks of historic stormwater releases of copper on fish populations, and 2) the risks of cumulative releases of landfill constituents on piscivorous wildlife. In the first case, stormwater data and simulation models were used to estimate concentrations of copper in river water. Copper concentrations were estimated to increase locally by less than 5% of ambient values with totals for individual storms for the past 5 years all less than the chronic criterion for the protection of aquatic organisms. In the second case, PCB concentrations in fish were estimated from measured sediment concentrations and exposures to piscivorous birds calculated based on described foraging patterns. In this case, exposures to the bird population were found to be negligible based on the reproduction toxicity of identified PCB compounds.

PWA118 Tiered Approach for Developing Ecological Risk-Based Remediation Objectives for RCRA Facilities. Morton, E.S.*; Monschein, E.S.; Tay, S.H.; Tetra Tech EM Inc. (Tetra Tech), Chicago, IL; Desmond, W.P., Ph.D., Tetra Tech, Dallas, TX; Yurk, J.J, U.S. Environmental Protection Agency Region 6, Multimedia Planning and Permit Division, Center for Combustion Science and Engineering. Risk-based corrective action at RCRA facilities is based on human health risk only – ecological risk is not commonly considered. However, protection of ecological receptors is the primary concern at many sites. This presentation describes a new approach for incorporating ecological risk assessment information within a tiered approach to determine remediation objectives. Available guidance strongly advocates the use of an iterative, tiered approach to performing ecological risk assessments that includes (I) a screening ecological risk assessment, followed by (II) the incorporation of as much site-specific information as possible to prepare more accurate estimates of potential ecological risks. When a tiered approach does not apply to an ecological risk assessment, the assessment can become lengthy, costly, and unfocused. Under the new approach, ecological Tier I screening values have been developed based on a conservative set of default assumptions. Specifically, Tier I values are largely based on results back-calculated from simplified food chain models developed for default, medium-specific target species. Tier II screening values, which incorporate site-specific information, are based on ecosystem-specific food web models developed in support of the U.S. Environmental Protection Agency Office of Solid Waste's multipathway screening level ecological risk assessment protocol. These food web models are used to back-calculate receptor- and ecosystem-specific remediation objectives. As a result of this effort, states will be able to apply both generic and site-specific ecological assessment values within the risk-based corrective action process to better assist facilities in conducting more timely and cost-effective ecological risk assessments and cleanups.

PWA119 Risk Based Decision Making: The East Fork Poplar Creek Case Study. Moore, D.R.J.*, Teed, R.S., The Cadmus Group, Ottawa, ON; Sample, B., Suter, G., Oak Ridge National Laboratory, Oak Ridge, TN; Parkhurst, B., The Cadmus Group, Laramie, WY. A probabilistic risk assessment revealed that methylmercury released from the U.S. DOE Y-12 weapons facility at Oak Ridge, Tennessee poses moderate risks to mink and kingfishers residing near the receiving waters of East Fork Poplar Creek. PCBs released from this facility pose severe risks to mink, but little risk to kingfishers. The objective of this study was to use a risk based decision making approach to select remedial cleanup levels for each of these contaminants. We conducted Monte Carlo simulations to estimate total daily intakes of each contaminant by mink (mercury and PCBs) and kingfishers (mercury only) for a range of exposure reduction scenarios. The resulting exposure distributions were then convolved with their respective dose-response curves to estimate post remediation risks. The results indicated that mercury levels would need to be reduced from current levels (mean=0.225 mg/L) to 0.02 mg/L to reduce risks to very low levels for both mink and kingfishers. If interested parties define acceptable risk as, for example, a 20% probability of 10% mortality, then mercury levels would need to be reduced to 0.12 mg/L. The PCBs analysis indicated that reducing water-borne exposures would produce only a modest reduction in risk to mink, because much of the current exposure is through terrestrial exposure pathways.

PWA120 Assessing Risk to Aquatic Biota Using Site Specific Assessment Tools for Multiple Exposure Pathways. Vertucci, F.A.*, Naddy, R.B., and Stubblefield, W.A. ENSR, Fort Collins, CO. Many risk assessments rely on ambient water quality criteria (AWQC) to evaluate potential risks to aquatic biota from contaminant exposure. The AWQC are not intended to consider risk from sediment or dietary exposure pathways and few assessments account for site-specific bioavailability of metals. Risks to aquatic biota from metals in the Clark Fork River (CFR), Montana were evaluated using assessment tools specific to each exposure medium; water, sediment, and diet. Each medium was evaluated using extensive site-specific exposure data and, when possible, effects criteria that reflect site metals bioavailiblity. Risk from exposures to water column metals was evaluated by comparing dissolved metals (As, Cd, Pb and Zn) concentrations to AWQC; for Cu the AWQC was adjusted based on site-specific water-effect- ratio studies (WER). Sediment metals exposure was evaluated using Sediment Quality Criteria (SQC) methods (i.e., SEM-AVS and sediment porewater summed as toxic units) and using bulk Sediment Effects Concentrations (SEC) derived from toxicity tests with sediments from the CFR watershed. Trout dietary exposures were addressed by comparing extensive benthic macroinvertebrate tissue data with literature-derived toxicity reference values (TRVs). Dissolved metals concentrations were generally lower than the AWQC for each metal (including Cu after WER adjustment). Metals in pore water and bulk sediments were below SQCs and SECs. Furthermore, benthic macroinvertebrate tissue concentrations did not exceed dietary TRVs. The results predict nominal risk to CFR aquatic biota. These findings suggest that WER adjusted AWQC, when available, should be used as a risk assessment tool and may prove sufficiently conservative to be protective of other exposure pathways.

PWA121 Factoring Exposure into Environmental Assessment: When is it Crucial for Decision-making? Tucker, M.E., Pittinger, C.A., Ruffing, C. J., Mihaich, E.; on behalf of the Ecological Risk Assessment Committee of the American Industrial Health Council. Fundamental to ecological risk assessment is the concept of comparing a stressor's exposure with an ecological effect. Evaluations which include only physical/chemical properties of stressors, lacking fate and exposure considerations, have traditionally been termed hazard assessments. Yet bright line distinctions between hazard and risk assessments are increasingly difficult to make. These hybrid or "risk-based" procedures combine elements of hazard assessment with surrogate measures of exposure (e.g., production volume, persistence or bioaccumulation estimates). It is possible to draw a continuum across the range and complexity of methods used to represent exposure in these assessments. Hybrid methods are increasingly being used to calculate metrics to enable chemical ranking or scoring, and are attractive in their simplicity. But environmental scientists of the American Industrial Health Council view inherently hazard-based environmental analyses as problematic when used as the sole criterion upon which environmental management decisions are based. The robustness of the exposure component in any "risk-based" environmental evaluation process should be commensurate with the ultimate decision at hand. The continued development of chemical management prioritization schemes, public "Right-to-Know" initiatives, harmonization of hazard classifications, and other programs requires clear definition of objectives and selection of appropriate tools. Communication of the key assumptions, uncertainty, and variability inherent in such approaches is critical. Representative programs in these areas will be assessed along a "hazard-risk continuum", and criteria will be recommended for sound and appropriate characterization of exposure in environmental evaluation systems.

PWA122 The development and implementation of the Policy for the Management of Toxic Substances of the Canadian Council of Ministers of the Environment (CCME). Lugsdin, T.L., Environment Canada, Hull, Quebec. The management and reduction of toxic substances in the environment has been identified as a national priority in Canada. The Canadian Council of Ministers of the Environment (CCME) endorsed a Policy for the Management of Toxic Substances (PMTS) in January 1998. The policy ensures that all jurisdictions work together to provide Canada with an integrated, cooperative approach for the sound management of toxic substances, with each government retaining its existing authorities but using them in a coordinated manner to achieve enhanced environmental results. The Policy provides Canadian jurisdictions with a common approach to establishing national priorities for toxic substances. The common approach includes mechanisms for substance identification, assessment of toxicity, and assigns toxic substances to one of two tracks (virtual elimination or life cycle management to prevent or minimize releses) depending on whether they result primarily from human activity and meet criteria for persistence, bioaccumulation and toxicity. The policy also calls for the development of Canada wide control strategies, implementation plans, and monitoring and public reporting provisions. The details of this multi-jurisdictional national policy, the successes and challenges encountered in it's development and an examination of its initial implementation will be presented.

PWA123 Exposure Assessment of Polychlorinated Dibenzo-p-dioxines and Polychlorinated Dibenzofurans by Multi-grid/Multi-media Fate Model. Higashino, H.*, Komai, T., Gamo, M., Yonezawa, Y., National Institute for Resources and Environment, AIST, MITI, JAPAN. A numerical simulation with a model of multi-grid/multi-media type was applied to exposure assessment of polychlorinated dibenzo-p-dioxines (PCDDs) and polychlorinated dibenzofurans (PCDFs). In Japan, since the greater part of waste is processed by incineration, a lot of small-scale waste incinerators, which is the main source of PCDDs/PCDFs, exist in the high population density region. Therefore, it is necessary to apply the model of multi-grid type to evaluate exposure from two or more sources. In order to evaluate various exposure routes, the model treats material transports between media (atmosphere, soil, sediment, river, underground water, and sea area). The model was applied to the area covering whole Kanto distinct (including Tokyo metropolitan area), where 200 or more incineration plants exist. The analytical domain with a scale of 276 x 224 km was discretized into 40 x 60 grids with space increments 4.6 x 5.6km. The gridded emission inventory of PCDDs/PCDFs in the analytical domain was compiled based on the published monitoring data on exhaust gas, the waste amount and the location of each municipal incineration. Exposure level at each grid was evaluated from the calculated concentration in each medium. The spatial distribution of the heavily polluted area was clarified. This results, combined with demographical and epidemiological data, must be an essential information for risk management of PCDDs/PCDFs.

PWA124 Dioxin Analyses for Ecological Risk Assessments: Scientific Approach and Practical Considerations. Henningsen, G.*, U.S.EPA, Denver, CO; Roy, R., U.S.FWS, Pasco, WA; Bradbury, S., U.S.EPA, Duluth, MN; Henry, T., U.S.EPA, Denver, CO; Hoff, D., U.S. EPA, Denver, CO. Successful evaluation of excessive risks to health caused by exposure to dioxins and related Ah-agonists (furans and co-planer PCBs) depends upon important biological and analytical factors. These factors are essential whenever evaluating risks posed by a class of potent chemicals with varying degrees of related toxicity (e.g., PAHs and PCBs). Use of Biological Technical Assistance Groups and outside experts on problems with complex stressors, such as dioxins, helps guide effective sampling and analyses that can address both remediation and restoration concerns for contaminated Superfund and other sites. Potent toxicants, like dioxins, challenge current analytical methods' capabilities for accuracy, precision, and low method detection limits with good QA/QC. To statistically quantitate potential risks to wildlife, method quantitation limits (MQL) should be about 2- to 10-fold lower than risk-based concentrations in media of concern. For 2,3,7,8-TCDD, the MQL often needs to be near or less than 1 ppt (pg/g). Varieties of field and laboratory analytical methods have advantages and disadvantages for certain applications; generally, use of bioassays with sensitive linear responses for total TEQ determinations, and GC/MS for specific congener determinations, are currently preferred. TEFs normalize Ah-agonist congeners to TEQs of 2,3,7,8-TCDD; use of WHO TEFs for terrestrial screening, and derivation of specific toxicity potency factors for more quantitative terrestrial risk and aquatic assessments, is recommended. Congener pattern analysis is sometimes indicated to help determine probable sources of Ah-agonists based on fingerprints of approximately 21 congeners. Sampling designs must be representative of receptors' exposure units (e.g., home-ranges), and suitable reference areas are essential for discerning site-related sources from prevalent natural background contamination.

PWA125 Establishing a Quality Assurance Program for Natural Attenuation. McInnis, D.L., Toxicological & Environmental Associates, Inc., Baton Rouge, LA; Droy, B. F., Ph.D., Toxicological & Environmental Associates, Inc., Baton Rouge, LA; Green, D., Department of Safety, Health and Environment (DSHE), United States Army – Aberdeen Proving Ground. Quality Assurance (QA), as it is applied to U.S. Environmental Protection Agency (EPA) programs such as RCRA and Superfund, is highly focused on the planning and implementation of data collection and analysis activities. These QA processes are critical to ensure the acquisition of high quality, useable and legally defensible data. However, the presence of valid analytical data alone does not necessarily ensure a correct interpretation of site contamination (e.g., the lateral and vertical extent, transport routes, and eventual fate of contaminants). To ensure the accuracy of site data interpretations, it is critical that QA processes be developed and applied to the data interpretation of environmental investigations. EPA's current QA process generally includes the following: establishing data quality objectives (DQOs) for the investigation; developing quality assurance project plans (QAPPs); and sampling and analysis plan (SAPs) that include and fully address EPA required elements; meeting established quality control (QC) requirements through the collection and analysis of field and laboratory audits. Unfortunately, these processes do not address data interpretation. The need for conclusions to be logical, complete and well documented is no different than the planning process in which DQOs are established in QAPPs are prepared. Standard and proven techniques and protocols, many developed and established through the EPA, can be applied to the interpretation of data. By implementing QA measures such as: 1) required elements for environmental technical reports (ETRs); 2) standard operating procedures (SOPs) for data interpretation techniques; and 30 checklists for assessing data interpretation activities, a greater degree of confidence in the overall investigation can be obtained. These QA procedures greatly benefit the decision-making process in selecting remedial alternatives. The use of a QA/QC program for natural attenuation data interpretation and applications will be presented.

PWA125a Debating Policy Alternatives for the Sustainable Management of Contaminants in the Environment. Harris, G.E.* and Gobas F.A.P.C. Simon Fraser University, Burnaby, B.C. Canada. The detection of exposure and effects of chemicals in the environment requires a continued role for contaminant management. The traditional management approach has relied on regulatory controls of industry to limit waste discharges. With limited government resources, social economic constraints, and increasing corporate stewardship, there are several alternative approaches being discussed to achieve environmental objectives. Pollution prevention is a key component of this approach and is being promoted by many of the new policy options. These include voluntary initiatives, the use of economic instruments, virtual elimination of specific chemicals, and determination of critical loads. Environmental limits are created by the finite capacity of ecological systems to remove chemicals from the environment and are discussed within the context of sustainable development. This paper develops criteria for the sustainable management of chemicals in the environment based on our understanding of chemical behaviour. The current policy alternatives are compared to the criteria. None of the alternatives meet all of the criteria which could lead to environmental conditions inconsistent with the goals of sustainable development and pollution prevention. An alternative framework is presented which incorporates our current understanding of chemicals in the environment and meets the larger social objectives of sustainable development.

PWA126 Isolation of accumulated contaminants associated with biological activity in fish: An example using bleached kraft mill effluent. Hewitt, L.M.*, Parrott, J.L., Servos, M.R. and Munkittrick, K.R., Environment Canada, Burlington ON. A major benefit derived from bioassay-directed chemical fractionation experiments conducted on complex environmental mixtures is the identification of biologically active compounds relevant to the environment. Fractionation experiments are usually directed using short term *in vivo* exposures or in vitro assays designed to rapidly provide an indication as to the presence/ absence of active chemicals. Concerns over the applicability of these bioassays to the dynamic exposures encountered in environmental situations relate primarily to the potential inability of these assays to account for the temporal fluctuations of multiple active components as well as long term accumulations through food webs. In an effort to address these concerns, our approach was to develop and apply methodology for fractionation experiments using liver tissues of wild fish. Methodology developed for isolating and identifying components in complex mixtures associated with mixed function oxidase (MFO) induction in fish was applied to liver tissues from white sucker (*Catostomus commersoni*) exposed to bleached kraft mill effluent and were directed by MFO induction in H4IIE cultures. Fish collected from a reference area as well as the effluent plume were caged in a reference stream and the effluent stream. Fractionation experiments indicate there are multiple classes of compounds associated with MFO induction which differ substantially between fish exposed historically and reference fish exposed short term. Differences in the estimated partitions coefficients of the chemical classes suggest both waterborne as well as food web routes of exposure in wild fish.

PWA127 Biochemical Defense Mechanisms against Pulp Mill Effluent-induced Damage in Estuarine Plants. Lytle, J.S.* and Lytle, T.F., Institute of Marine Sciences, Gulf Coast Research Laboratory, The University of Southern Mississippi, Ocean Springs, MS Phenolic compounds are the chief constituents of paper mill wastes. Those aquatic plants that survive in these niches most likely have protective mechanisms which detoxifies the damaging effects of these effluents allowing them to adapt to their environment. Peroxidase is a major enzyme system catalyzing oxidative metabolism of xenobiotics in plants. These enzymes are ubiquitous in plants and may play a major role in the ability of the plant to tolerate phenolic wastes. Other defense mechanisms such as antioxidants may also play a role in plant sensitivity to xenobiotics. Plants are known to activate these defense mechanisms in response to environmental stresses. A study was carried out to compare peroxidase activity and antioxidant responses in *Spartina alterniflora* and *Juncus roemerianus* growing in the vicinity of a pulp mill with these species growing in a more pristine area. Peroxidase activity in *S. alterniflora* growing near the pulp mill was 5X that of the same species growing at a clean site, whereas peroxidase activity in *J. roemerianus* showed only small differences between paper mill and pristine sites. Ascorbate levels, though, were elevated in both species collected from the vicinity of the paper mill compared to those at the clean site and glutathione levels were depressed in both species growing near the paper mill as compared to those growing at the more pristine site. Increased oxygen radical generation and lipid peroxidation have been suggested to be responsible for the toxic actions of a wide range of compounds in plants. However, if the plant has sufficient antioxidants and/or enzymes that can metabolize the toxic compounds, the plant species which canthe availability of antioxidants.

PWA128 Comparison Between the Effects of the Phytosterol β-Sitosterol and Pulp and Paper Mill Effluents on Sexually Immature Rainbow Trout. Tremblay, L.*, G. Van Der Kraak, Department of Zoology, University of Guelph, Guelph, ON, Canada. Smaller gonads, delayed sexual maturity and depression of circulating sex steroids have been reported in feral fish exposed to pulp and paper mill effluents. The plant sterol β-sitosterol, found in significant amounts in pulp and paper mill effluents has been shown to cause reductions in sex steroid secretion in goldfish may contribute to some of these effects. In this study, the effects of 17β-estradiol (E_2), two sources of β-sitosterol and two Canadian bleached mill effluents (BME) on sexually immature rainbow trout were investigated in a 21 d *in vivo* experiment. Both β-sitosterol sources exerted similar effects as did BME. Plasma vitellogenin (Vtg) was significantly induced by BME and β-sitosterol suggesting an estrogenic response. β-Sitosterol had no effect on testosterone levels while one of the BME significantly reduced testosterone. Pregnenolone was significantly reduced by β-sitosterol and the two BME but not by E_2 suggesting that this was not an estrogenic effect. Total plasma cholesterol was reduced by β-sitosterol but not significantly by BME. Feral female white sucker exposed to BME also showed significantly reduced plasma cholesterol when compared to females from a reference site. Mixed-function oxygenase (MFO) activity was significantly induced by β-sitosterol and BME exposed fish. Both BME and β-sitosterol were shown to exert biological responses through more than one mechanism of action. These studies suggest that β-sitosterol could contribute to some of the biological effects caused by pulp and paper mill effluents.

PWA129 Sex Inversion in Fathead Minnows, *Pimephales promelas*, Exposed Since Embryos to Pulp and Paper Effluent. II: A proposed pathogen/chemical pollution etiology for masculinization /feminization events in fish. Anderson, P.D.*, Lauzon, S., Ruby S., Brousseau P., Concordia University, Montreal, Spear P. TOXEN, UQAM, Montreal. Various tissues of 10-month old minnows were examined histologically for lesions related to masculinization of their populations from exposure to BKME. Lesions were consistent with a pathogenic infestation. Internal pathology correlated with overt signs of microbial infection. Gonads also showed lesions. Some testis were suppressed in development. Other individual's testes were highly developed, active in spermatogenesis and contained evidence of vestigial primary follicles. Ovaries displayed atretic and/or highly vacuolated follicles and a high incidence of "brown bodies". Stroma tissue appeared depleted. It follows that thecal layers may have been damaged at a critical point in the development of primary follicles. he thecal layer provides the aromatase enzyme that converts testosterone from follicle cells to oestrogen. With thecal tissue loss, testosterone levels increase and induce primordial germ cell tissue to evolve as testis. The result is masculinization of gonads and somatic tissues of an otherwise genotypic female. These masculinized females were found to grow large and to be aggressive breeders thus increasing the likelihood of females in the next generation. This may lead to a feminization of fish populations. In contrast, it appeared that pathogenic damage to stroma tissue in developing male gonads results in loss of interstitial cells and thereby the production of testosterone. Consequently, testicular development is suppressed in true males resulting in a decrease in the next generation of genotypic male. If this theory of sex inversion proves valid, a refocus from hormonal mimics and enzyme inhibitors as causal agents to carbon loading and immunosuppressants in effluents is necessary.

PWA130 The Comparative Effect of Bleaching and Non-bleaching Pulp Mill Effluents on the Antioxidant Defenses of Tilapia, *Oreochromis Niloticus*. Wilhelm Filho, D.;Testa, C.P.; Tribess T.B.; Torres, M.A.; Soares, C.H.; Pedrosa, R.C.*; Baptista, I. Universidade Federal de Santa Catarina, Brazil. The effect of sub-chronic treatment of bleaching and non-bleaching pulp mill effluents on the liver of *Oreochromis niloticus* was monitored during four weeks concerning antioxidant defenses (AD), O_2 consumption, and cellular damage (TBARS levels). Liver O_2 consumption were essentially the same in fish exposed to both treated and non-treated effluents of both processes. The cellular damage was significantly increased ($P<0.05$) in fish exposed to the different types of bleaching effluents (E1, final treated and final non-treated effluent) and also to the final non-treated effluent of the non-bleaching process, but remained unaltered in fish exposed to non-treated effluent of the non-bleaching process. Nevertheless, these changes were followed by an increase of glutathione S-transferase activities in fish exposed to bleaching and non-bleaching effluents. Superoxide dismutase activities did not change, but catalase activities were enhanced in fish exposed to both effluents. Irrespective of the type or stage of the effluent, total glutathione concentrations gradually decreased during the exposure period. In conclusion, the antioxidant defenses of tilapia exposed to the bleaching and non-bleaching processes indicates that the industrial treatment is not sufficient to avoid an oxidative stress in fish.

PWA131 Cytochrome P4501A and Glutathione S-Transferase Induction in Tilapia *(Oreochromis Niloticus)* Exposed to Bleaching and Non-bleaching Eucalyptus Pulp Mill Effluent from Brazilian Pulp-and-Paper Industry. Pedrosa R.C.*, Geremias R., Locatelli C., Soares C.H.L, Baptista I.E., Wilhelm Filho D., Federal University of Santa Catarina - CCB, SC, Brazil. Cytochrome P4501A and glutathione S-transferase are the enzymatic systems of phase I and II biotransformation that can be induced by polyhalogenated aromatic hydrocarbons (PAH). This induction is frequently assessed as ethoxyresorufin-O-deethylase (EROD) and GST activities. In order to better understand the contribution of bleaching and non-bleaching pulp processes in the toxicity of pulp mill effluent, juvenile tilapias (~l00 g, n = 5) were held in 52 L tanks and exposed to two eucalyptus pulp mill effluents (dilution: 1/150) during 4 weeks. At the end of the exposure period, fish were killed and the livers rapidly removed. Then the EROD and GST activities were determined in microsomal and citosolyc fractions. A significant difference ($P<0,05$) between CYP45O1A and glutathione S-transferase expressions in fishes exposed to the two effluents was obtained. The induction of these enzymatic systems were stronger in bleaching pulp mill. However, non bleaching effluent sustained the MFO and GST induction, but in a smaller level. Using the biotransformation enzyme induction as a toxicity bioindicator it is possible to conclude that non bleaching process of pulp-and-paper brazilian industry reduced the toxicity of effluent but do not remove completely the toxicity compounds.

PWA132 Effects of bleached and unbleached hardwood pulp effluents on liver and gill morphology in Tilapia (*Oreochromis niloticus*). Soares, C. H. L. *; Baptista, I. E. *; Whilhelm Filho, D.** and Pedrosa, R. C.* Departamento de Bioquímica* and Departamento de Ecologia & Zoologia**, Centro de Ciências Biológicas, Universidade Federal de Santa Catarina, Cx.P. 5079 CEP 88040-970 Florianópolis, SC, Brazil. The effluents from paper and pulp industry contain a wide range of organic materials, such as chlorophenols and other lignin derivative compounds. The composition of these effluents varies considerably depending of wood species used, cooking conditions and bleaching (or not) sequences employed, among the others factors. And so, sometimes, it is difficult to verify a clear cause-and-effect relationship between a particular effluent component and biological responses. From an environmental standpoint, it is of great interest either to determine the nature and amount of these organic materials as well as their toxicity effects on aquatic organisms. The histopathological effects of unbleached and bleached pulp effluents were studied under experimental conditions. Adult tilapia (*Oreochromis niloticus*) weighing about 70 g, acclimatized for two months prior to utilization, were employed under sublethal toxicity conditions. Twenty five fish were kept in 310 l aerated plastic tanks containing 250 l of a 1/150-diluted effluent with the following physicochemical characteristics, pH = 6.8-7.0 ; dissolved oxygen = 8.0-10.0 mg/l; water conductivity = 217 mS/m, and the temperature at 25.0 ± 1.0 °C. The water was daily changed and a light:dark cycle of 13:11 h was used. One control group, treated identically, was kept in fresh water (conductivity = 7.1 mS/m). Five fish from each group were weekly examined for gill and liver tissue lesions by conventional histological methods during five weeks. In this period, similar effects for both effluents were observed, however, occuring with different intensities. From slight (second week) to severe (fourth-fifth weeks) hyperplasia of secondary lamellae of the gills with a consequent fusion lamellae were found for both types of effluent. Hyperplasia in the hepatic tissue, which exhibited areas of vacuolation, were also found in the effluent-exposed fish. Morphological signs of intracelular content loss in addition to cells with displaced nuclei or surrounded by vacuoles could be observed.

PWA133 Development of a North American research strategy to assess potential reproductive effects of pulp mill effluents on fish – focus on hormone-mediated effects and assessment. Rodgers, J.H.*, Clemson University, Pendleton, SC; Fisher, R.P., National Council for Air and Stream Improvement, Research Triangle Park, NC; Festa, J.L., American Forest and Paper Association, Washington, D.C. This workshop developed a strategy that could be employed by the sponsors (NCASI and AF&PA) in conducting and sponsoring research on potential effects of pulp and paper mill effluents on fish reproduction in effluent receiving waters, and on possible mechanisms of such effects. The workshop was prompted by reports in scientific literature that indicated reproductive system changes in fish downstream from some pulp mill effluent discharge points. The content and format of the workshop included: 1) sixteen scientists from the United States and Canada representing academia, government and industry, 2) presentations followed by discussion on each, 3) development of a research strategy by two independent working groups, and 4) plenary development of a consensus research strategy in response to workshop goals. This poster will present the research strategy that emerged from the diverse views on the issues regarding existing experimental data and research findings.

PWA134 Periphyton and sediment bioassessment as indicators of the effect of a coastal pulp mill wastewater. Dantin, D.D.*, U.S. EPA, Gulf Breeze, FL; Lewis, M.A., U.S. EPA, Gulf Breeze, FL; Foss, S.S., U.S. EPA, Gulf Breeze, FL. A two year study was conducted near Port St. Joe, Florida, in a coastal transportation canal and bay receiving combined municipal and pulp mill wastewater. The objective of the study was to determine the effectiveness of periphyton analysis techniques and sediment toxicity as indicators of environmental stress. Periphyton were colonized for 3 weeks in-situ at 5 locations above and below the outfall. Sediments were analyzed for toxicity using epibenthic grass shrimp and mysids. Despite decreases in the photic zone, significant increases in biomass and associated pigment production occurred below the discharge point. The percent increase in biomass downstream was as much as 500%, compared to periphyton colonized above the outfall. Sediments were found to be toxic to the grass shrimp but not to mysids. These effects of the effluent decreased progressively downstream of the outfall, indicating that the impact was site-specific. These field-derived effects were not predicted from the standard single-species toxicity tests conducted with and alga, invertebrate, fish, and the wastewater. It was concluded that the combination of single species toxicity tests and analysis of the periphyton and sediment toxicity were needed to define the effects of this wastewater in an estuarine environment.

PWA135 Comparison of Air Deposition of Mercury with Concentrations in Fish. Keating, F.J.*, Cocca, P.A., U.S. EPA, Washington, DC. The U.S. EPA has assembled a national data base of mercury concentrations in fish from state monitoring conducted during 1990-1995. The database includes sampling of a variety of species from over 4500 discrete locations within approximately 3000 discrete water bodies. The spatial distribution of sampling represents broad coverage for most states in the eastern half of the country. The EPA has also produced a national coverage of estimated total wet and dry air deposition of mercury for a combination of 1990 U.S. emissions and global background using the Regulatory Model for Aerosols and Deposition (REMSAD). Estimated dry and wet depostion amounts for total mercury vary from less than 5 g/km^2 to over 100 g/km^2 in the eastern half of the U.S. For this presentation, model output for wet deposition will be compared to wet deposition monitoring from a regional test area as an indicator of overall model validity. Using GIS overlay analysis to relate the point coverage of mercury in fish to the polygon coverage of mercury deposition, statistical analyses will be performed to show the degree of national and regional correlation, and differences in frequency distributions of fish tissue concentrations by estimated deposition category. The presentation will highlight the many mitigating factors and limitations inherent in such a comparison. To the extent possible, the effect of relevant water quality factors (e.g., pH) on fish concentration will be examined relative to the correlation with deposition. Questions to be addressed include "To what extent can air deposition of mercury be directly related to environmental exposure to mercury in fish?" and "How large a role do site specific environmental chemistry, other mercury sources, and other factors likely play?".

PWA136 Mercury Contamination in El-Mex Bay, Alexandria, Egypt. Khan, A.A.*, D.A. Hinckley, EA Engineering, Science and Technology, Sparks, MD, USA. The study was conducted as part of the Mediterranean Sea Environmental Characterization Program (MSECP) which supported an Environmental Assessment (EA) commissioned by the United States Agency for International Development (USAID). The objective of the study was to evaluate mercury contamination in water, sediment and biota from El-Mex Bay. The bay, located on the western side of Alexandria City, represents a highly eutrophic and contaminated coastal ecosystem. Previous studies have shown that the bay receives significant amounts of mercury from the discharge of a chloroalkali plant. Water, sediment and biota samples were collected during October and November 1996 from selected locations in the bay and analyzed for total mercury. Fish and macroinvertebrates that were popular food items were selected for biota analyses since these would provide a pathway for mercury transfer to humans. Whole body samples were analyzed since most fish are generally consumed whole by local fisherman. Analytical results indicated elevated mercury concentrations in water and sediments samples from several locations in the bay. Tissue mercury concentrations ranged from 0.31 to 2.30 mg/kg wet weight. Four of the seven species had whole body tissue concentrations in excess of 1 mg/kg, the edible fish tissue mercury limit established by the World Health Organization (WHO) and the Food and Drug Administration (FDA).

PWA137 Distribution of Methyl Mercury in Sediments from Korean Coastal Areas. Lee, K.T.*, Seoul National University, Seoul, Korea; Kannan, K., Michigan State University, Michigan, USA; Shim, W.J., Korea Ocean Research and Development Institute, Seoul, Korea; Koh, C.H., Seoul National University, Korea. To elucidate contamination levels and distribution of methyl mercury in coastal areas of Korea, methyl mercury concentrations were determined in 126 sediments collected from Kyeonggi Bay, Namyang Bay, Chinhae Bay, and Lake Shihwa, during 1995 - 1996 were determined by cold vapor atomic fluorescence spectrometry. Concentrations of methyl mercury in Kyeonggi Bay, Namyang Bay, Chinhae Bay, and Lake Shihwa were 274 ± 990, 108 ± 24, 294 ± 342, and 1080 ± 760 pg/g, respectively. Methyl mercury concentrations in sediments were significantly correlated with total organic carbon and total sulfur contents, but were independent of mud contents and mean grain size. Apparent gradient was observed in methyl mercury concentrations in a concentration-contour map around three sites (S5, S6, and S10) in Lake Shihwa, implying that methyl mercury accumulation in sediments may be influenced by wastes from industrial complexes. Relatively higher concentrations of methyl mercury at surface and 10 cm in depth in Chinhae Bay core sediments may be responsible for higher rate of methylation process by active sulfate reducing bacteria or implicative of greater concentration of mercury available to microbes for methylation.

PWA138 Comparison of elements in bottlenose dolphins stranded on the beaches of Texas and Florida in the Gulf of Mexico. Meador, J.P.*, Ernest, D., Tilbury, K., Stein, J.E., NOAA Fisheries, Seattle, WA; Hohn, A.A. NOAA Fisheries, Beaufort, North Carolina. We analyzed tissue samples from bottlenose dolphins (*Tursiops truncatus*) that had stranded on beaches in Texas and Florida over a one-year period starting in September 1991. The concentrations of ten elements plus methyl mercury (MeHg) were determined in brain, kidney, and liver, and we examined these results for differences based upon age, site, sex, and tissue type. A strong inverse relationship between total mercury (Hg) and the percentage that was methyl mercury (MeHg) was found in liver, kidney, and brain tissue, presumably due to demethylation of MeHg. A threshold concentration was found for total Hg in brain tissue, indicating that most Hg was present as MeHg up to about 8 years of age. Increases in total Hg after this age were accompanied by an increase in the ratio of total Hg to MeHg, indicating demethylation. Strong relationships were found between total Hg in liver and age and between total Hg and selenium in liver, which have been observed before in many fish and squid-eating marine mammals. The only difference based on sex of the animals was observed for MeHg, which was higher in females and contrary to the pattern often observed for organic contaminants. Several elements (copper, Hg, lead, zinc) exhibited intersite differences, which were not consistent. Bottlenose dolphin from Florida exhibited the highest levels of MeHg and total Hg, while animals from Texas exhibited the highest levels of lead, copper, and zinc. The essential elements copper and zinc were expected to be the same for the Texas and Florida animals; however, observed differences may indicate population differences in basic physiological levels, dietary intake, or health status.

PWA139 Development of National Default Bioaccumulation Factors (BAFs) for Mercury. Linton, T.K.*, Clement, W.H., McIntyre, D.O., Great Lakes Environmental Center, Columbus, OH; Sappington, K., U.S. EPA, Washington, DC. Bioaccumulation factors (BAFs) are currently being used by the U.S. Environmental Protection Agency (U.S. EPA) to develop water quality criteria for the protection of wildlife and human health. The work presented in this poster is an extension of the work conducted by U.S.

EPA and presented in the Mercury Report to Congress (December 1997). The goal of this work was to calculate national default BAFs for methylmercury in freshwater (lentic/lotic) and coastal (estuarine) environments and to determine if there are differences in the bioaccumulation of mercury between these different systems. A literature search strategy was devised to obtain all pertinent literature since the early 1990's (i.e. since the employment of "ultra clean" techniques). Applicable data were extracted and used to derive default BAFs for methylmercury. Calculations were based on the following approaches: 1) direct method - quotient of methylmercury in aquatic animals (trophic levels 2, 3, and 4) to dissolved methylmercury in the water column, 2) indirect method - estimated as above but using a conversion factor to convert total dissolved mercury to dissolved methylmercury in the water column, and 3) a modification of the approach outlined in the Great Lakes Water Quality Initiative Technical Support Document (EPA-820-8-95-005). BAF values for each aquatic system (lentic, lotic, estuarine) were determined and subsequently compared. Methylmercury and total mercury forms were evaluated to determine which forms of mercury result in the best prediction of mercury accumulation. In addition to the BAFs, the partitioning of total mercury and methylmercury between the dissolved and particulate phases in the different systems were calculated using two approaches (direct dissolved fraction and derivation from K_D and TSS). Data gaps were identified and the uncertainty associated with each default BAF estimate calculated.

PWA140 Bioaccumulation of Mercury in Catfish from the Ismailia Canal of Egypt. El-Boushy, M. E. and Abd-Alla, O. A., Suez Canal University, Egypt and Khan*, A. T. School of Veterinary Medicine, Tuskegee University, AL. Catfish samples were collected from different locations of Ismailia Canal to evaluate the levels of contaminants. Catfish liver, kidney and muscle samples were analyzed for mercury by cold vapor atomic absorption spectrophotometer. The mean levels of mercury in the livers were 20.00, 15.70, 8.57, 5.90, and 1.70 ppm at Mostord, Abozabal, Belbeas, El-Abebsa and Ismailia, respectively. Mercury level in the kidneys were 29.80, 21.60, 11.50, 7.10 and 3.60 at the same locations, respectively. The mean levels of mercury in the muscles were 0.71, 0.62, 0.38, 0.27, and 0.11 ppm at the same locations, respectively. The level of mercury was highest in the kidney and lowest in the muscle at all locations. This pattern of distribution is similar to that expected with mammals. The level of mercury was highest in Mostord catfish while lowest in Ismailia catfish in comparison to other locations. These data indicate that the Ismailia Canal at Mostord is highly polluted with mercury.

PWA141 Mercury Accumulation in Fish and Bird Prey Items From a Texas Gulf Coast Estuary. Robinson, S.C.*, Parametrix Inc, Kirkland, Washington; Quast, W., Hammons, D., and Young, H., Parametrix Inc., Houston, Texas; Gribben, K., Aluminum Company of America, Pittsburgh, Pennsylvania. Lavaca Bay is a shallow Texas gulf coast estuary and an ecologically diverse habitat providing foraging and breeding grounds for a wide variety of species. Elevated concentrations of mercury have been measured in Bay sediments, and in the tissues of finfish (red drum, *Sciaenops occelatus*), fish-eating birds (Forster's tern, *Sterna forsteri*) and invertebrate-eating birds (Willet, *Catoptrophorus semipalmatus*) occurring at the Site. A study was conducted to concomitantly measure the concentrations of mercury (total, methyl) in sediments and in the tissues of numerous invertebrates and small fish representing prey for these fish and birds. Sediment and prey were collected from three different habitat types occcurring in the Bay. Though primarily used as input to the wildlife and aquatic ecological risk assessment dose calculations, *a posteriori* evaluations of the data were also performed to determine if statistical relationships existed between the co-located sediment and tissue data, and to ascertain the role (if any) of habitat type on mercury accumulation potential. Biota-sediment accumulation factors (BSAFs) were also developed for non-normalized or organic carbon normalized sediment. Results suggest a strong statistical correlation between concentrations in sediment and tissue for most species based on a linear model. Findings also suggest that habitat may play a role in mercury accumulation for the limited number of species for which cross habitat data was available; though limited sample size may play a part in the observed results. Estimated biota-sediment accumulation factors were found to be highly variable.

PWA142 Pathways of Accumulation of Mercury in an Estuarine Food Web. G.M. Pastershank* and D. Lean. University of Ottawa, Ottawa, ON, K1N 6N5. The pathways by which methyl- and total-mercury is accumulated in estuary benthic and pelagic organisms is currently under investigation. Water, suspended particulate matter, depositional sediments, primary producers, invertebrates, fishes, and double crested cormorant eggs collected from the Miramichi River Estuary were analyzed for methyl- and total-mercury (by CVAAS) and carbon and nitrogen isotopes (on a CE Instrument EA-110, and Conflow II Interface with a Finnighan Mat Delta^{PLUS} IRMS). Carbon and nitrogen isotopes helped trace sources of energy and identify trophic positions of estuarine organisms, respectively. Freshwater, estuarine, and marine primary producers occupy the lowest trophic positions, followed by zooplankton, filter-feeders and other estuary invertebrates. Top trophic level organisms (δ15N of 11.5 to 15.5‰) consisted of mummichugs, sticklebacks, eels, gaspereau, herring, and juvenile and adult tomcod, winter flounders, rainbow smelt, and striped bass. Predictive relationships between carbon and nitrogen isotopes and methyl- and total-mercury for the Miramichi River Estuary food web will be presented.

PWA143 Confidence Intervals for Tolerance Data in Toxicity Testing for Probabilistic Risk Assessment. Farrar, D.B.*; Fite, E.C.; Frankenberry, M.J., U.S. EPA, Washington, DC. A Monte Carlo method was employed for calculating confidence intervals for fractional response data at a range of given doses. The method was used with data from acute toxicity testing in a variety of terrestrial species. One and two sided intervals were developed to produce a matrix of response values and corresponding confidence levels per range of doses. This nomogram will aid researchers and regulators in identifying potential areas for further testing.

PWA144 A Probabilistic Risk Assessment of the Effects of Methylmercury and PCBs on Mink and Kingfishers Along East Fork Poplar Creek, Oak Ridge, Tennessee. Moore, D.R.J.*, Teed, R.S., The Cadmus Group, Ottawa, ON; Sample, B., Suter, G., Oak Ridge National Laboratory, Oak Ridge, TN; Parkhurst, B., The Cadmus Group, Laramie, WY. Over 50 years of operations, storage and disposal of wastes from the U.S. DOE Y-12 nuclear weapons facility at Oak Ridge, Tennessee has resulted in the contamination of water, sediment, biota and floodplain soils of East Fork Poplar Creek. A preliminary assessment revealed that methylmercury and PCBs were the contaminants of most concern and because these contaminants are persistent, accumulate in tissues and biomagnify up the food chain, piscivorous wildlife are the biota at greatest risk of exposure. The objective of this study was to estimate the risks posed by methylmercury and PCBs to two piscivorous species, mink and kingfishers. We conducted Monte Carlo simulations to estimate total daily intakes of each contaminant by each species and then convolved the resulting distributions with their respective dose-response curves to estimate risks. The results indicated that methylmercury poses a moderate risk to female mink (26% probability of at least 15% mortality) and kingfishers (50% probability of at least an 11 to 28% decline in fecundity depending on location). PCBs pose a very serious risk to mink (51% probability of at least a 50% decline in reproductive fecundity), a species known to be particularly sensitive to the effects of organochlorine substances, but little risk to kingfishers (<5% probability of a decline in reproductive fecundity greater than 10% at any location).

PWA145 Aggregate Residential Exposure Model for Pesticides. Sinha P*, Daly HE, Cohen JT, Bowers TS. Gradient Corporation, Cambridge, MA. The Food Quality Protection Act (FQPA) of 1996 was enacted to amend both the Federal Insecticide, Fungicide, and Rodenticide Act and the Federal Food, Drug, and Cosmetics Act. The FQPA defines 'safe' tolerance for pesticide chemical residues as "reasonable certainty that no harm will result from aggregate exposure to the pesticide chemical residue, including all anticipated dietary exposures and all other exposures for which there is reliable information." Therefore, under the FQPA, the EPA must assess potential exposure to pesticides from the multiple sources: dietary, drinking water, occupational, and residential sources. Additionally, for each source, analysis of exposure through dermal contact, ingestion, and inhalation are required for the assessment to be considered 'aggregate' and therefore acceptable. In another report entitled *Science and Judgment in Risk Assessment,* the

National Research Council recommends that both uncertainty and variability are accounted for when performing risk assessments. We have developed a single chemical, aggregate model to assess effects from both chronic and acute residential pesticide exposure using Monte Carlo methods to account for uncertainty and variability. Results from chronic exposure scenarios show a range of intakes for individuals in a population, due to variability in model parameters and a range of mean intakes over populations due to model parameter uncertainty. Our case study suggests that pesticide exposure (mg/kg-day) over a lifetime is age-dependent with higher intake occurring at younger ages. The model has the advantage of being flexible *i.e.*, input variables can easily be tailored to describe any exposure scenario, and analyze any pesticide. While the model addresses residential pesticide exposures only, the methodology can easily be adapted to address other exposure components (*i.e.*, dietary, drinking water, and occupational) to create a complete aggregate exposure assessment.

PWA146 Probabilistic assessment of risks to birds from pesticide sprays in orchards. Hart, A., Central Science Laboratory, York, UK. This paper reports one of two case studies conducted to evaluate the potential of probabilistic approaches to risk assessment for pesticides. This study focussed on risks to birds from the use of chlorpyrifos, pirimiphos-methyl and carbaryl in UK apple orchards. Conventional, deterministic methods of risk assessment based on worst-case assumptions indicated a high potential risk for some species, as birds which feed exclusively on insects contaminated by spraying would obtain a lethal dose in less than one day's intake. However, in practice very few birds obtain all their food from sprayed orchards. Radio-tracking studies were conducted for 4 species in a representative range of orchards. The 4 species showed contrasting patterns of habitat use, but in each species most individuals spent very little time in the orchard centre. The 4 species take different types of food which carry different levels of pesticide residues. Furthermore, the toxicity of each pesticide varies widely between species. When probabilistic methods were used to take account of these sources of variation it was concluded that mortality was possible, but would affect only a small proportion of individuals in the most sensitive species. This result is consistent with the results of field studies which found only limited and transient depression of plasma cholinestase activity in birds captured in orchards after spraying of the 3 pesticides. By contrast, the conventional, deterministic approach would have suggested a much higher level of risk. The future role of probabilistic methods in regulatory risk assessment is discussed.

PWA147 Probabilistic assessment of risks to birds from pesticide treated seeds. Hart, A., Central Science Laboratory, York, UK. This paper reports one of two case studies conducted to evaluate the potential of probabilistic approaches to risk assessment for pesticides. This study focussed on risks to birds from wheat seed treated with the OP insecticide fonofos. Previous field and laboratory studies on fonofos and pigeons had quantified a range of factors which are important in determining exposure. These include the area sown with treated seed, residue decay, the density of exposed seed, frequency of seed spills, attractiveness of sown fields, the availability of alternative foods, the rate of feeding, and the ability of birds to selectively avoid treated seed. Also, the toxicity of fonofos varies widely between species. Conventional, deterministic methods of risk assessment based on worst-case assumptions indicated a very high potential risk, as a pigeon feeding exclusively on treated seed would obtain a lethal dose of fonofos in a fraction of its daily intake. When probabilistic methods were used to take account of variation in the factors affecting risk, it was concluded that lethal poisoning will occur only when the feeding rate of pigeons, and residues of fonofos on the seed, are both very close to the upper limit of their range of variation. This is likely to be very rare when treated seed is sown efficiently, but more frequent when seed is accidentally spilled on the soil surface. The results of the probabilistic approach agreed very well with the actual frequency of reported poisoning incidents in the UK involving pigeons and fonofos (less than one per year), whereas the conventional deterministic approach over-estimated risk by a very substantial margin.

PWA148 Probabilistic Aquatic Risk Assessment of Pyrethroids: 1. Distributional Analyses of Laboratory Aquatic Toxicity Data. Solomon, K.R.*, University of Guelph, Guelph, ON; Giddings, J.M., Springborn Laboratories, Wareham, MA; Maund, S.J. Zeneca Agrochemicals, Berkshire, UK. On behalf of the Pyrethroid Working Group (PWG). It has been suggested that the higher tiers of pesticide risk assessment should contain a probabilistic element, since this enables the range of exposure and effect concentrations to be explored, and an estimate of the probability of impacts occurring to be derived. For the synthetic pyrethroids, there is a wealth of laboratory toxicity data on aquatic organisms which can be used to develop probabilistic distributions of effect concentrations. These data are reviewed here, and we describe the application of distributional analysis and its use in effects characterisation. The key findings were as follows:

- There were distinct differences in the responses of arthropods, fish, and aquatic plants as would be expected from the mechanism of action of the compounds.
- The distributions of toxicity were similar for all compounds, and for the majority of compounds were broadly ranked as would have been predicted from their insecticidal activity.
- Cypermethrin has often been used as a representative of the pyrethroid class in previous regulatory discussions, and these data confirmed that a risk assessment based on cypermethrin would be applicable to other pyrethroids.

If the inherent mode of action is duly considered, probabilistic approaches to effects characterisation appear to offer a promising approach for the higher tiers of risk assessment for pesticides.

PWA149 Probabilistic Aquatic Risk Assessment of Pyrethroids: 2. Effects Characterisation From Aquatic Mesocosm and Field Studies Giddings, J.M.*, Springborn Laboratories, Wareham, MA; Solomon, K.R., University of Guelph, Guelph, ON; Maund, S.J., Zeneca Agrochemicals, Berkshire, UK. On behalf of the Pyrethroid Working Group (PWG). The ecological relevance of effects characterisations based on laboratory data can be evaluated with data generated under field conditions. For pyrethroids, a considerable number of field studies have been conducted. Detailed review of a range of field studies on cypermethrin and esfenvalerate enabled a number of conclusions to be reached:

- Results revealed a trend in sensitivity from amphipods, isopods, midges, mayflies, copepods, and cladocerans (most sensitive) to fish, snails, oligochaetes, and rotifers (least sensitive).
- With few exceptions, populations affected by pyrethroids in the mesocosms recovered to normal levels before the end of the year of exposure; most populations recovered within weeks.
- The effects of pyrethroids observed in the mesocosms were correlated with maximum pyrethroid concentrations in the water, not with concentrations in the sediments.
- Effects did not appear to increase with cumulative exposure from repeated pyrethroid applications.
- Direct effects on fish were only observed at very high concentrations close to the water solubility. Indirect effects on fish were not observed.

The Lowest Observed Effect Concentration (LOECs) for cypermethrin based on ecosystem response was judged to be 100 ng/l, substantially higher than the 10th centile of the laboratory-based toxicity distribution. This difference in effect concentrations is most likely due to the rapid dissipation of pyrethroids and ecological factors which mitigate effects on exposed populations in the field.

PWA150 Probabilistic Aquatic Risk Assessment of Pyrethroids: 3. Remote Sensing/GIS Approaches for Mississippi Cotton. Hendley, P*, Zeneca Ag Products, Richmond, CA; Travis, K.Z., Zeneca Agrochemicals, Bracknell, UK; Holmes, C., Henriksen, E., Kay, S., Compliance Services International, Tacoma, WA. On behalf of the Pyrethroid Working Group (PWG). This study was designed to quantify key "real-world" factors that influence the potential for static water bodies to be exposed to pyrethroid insecticides from use in cotton in Yazoo County, Mississippi (one of the most extreme US cotton-growing counties for pyrethroid runoff and drift). Using a Geographic Information System (GIS), a Land-Use/Land Cover (LU/LC) classification from LANDSATTM imagery was combined with other digital data sets to characterize the types of water body and analyze their proximity to cotton fields. Soils and elevation data also allowed assessment of runoff potential. High resolution aerial photography was used to investigate the detailed composition and width of buffers between agriculture and water. Highlights from the results include:

- Cotton fields are mostly distant from static water bodies (68% of all ponds had no cotton within 360 meters; 92% of ponds had no cotton within 60 meters).
- For most ponds, the spatial distribution of cotton in the margins means that only a small proportion of wind directions will cause drift towards the ponds.
- Cotton is selectively cropped on flatter land; standard properties assumed for modeling runoff in Yazoo County over-emphasize the runoff potential.
- Typically, there are physical buffers (average width 25 m) between cotton and ponds which will impede drift and runoff. In the majority of cases, these buffers consisted of dense stands of trees.

PWA151 Probabilistic Aquatic Risk Assessment of Pyrethroids: 4. Landscape-Level Exposure Characterisation. Travis, K.Z*., Zeneca Agrochemicals, Bracknell, UK; Hendley, P., Zeneca Ag Products, Richmond, CA; Ritter, A.M., Waterborne Environmental Inc., Leesburg, VA. On behalf of the Pyrethroid Working Group (PWG). Under the current US-EPA pesticide risk assessment scheme, the established scenarios used to determine exposure concentrations contain relatively conservative assumptions about compound use and fate, and the agronomic and environmental conditions. For higher tier assessments, there is a need to be able to account for the complexities of spatial relationships between ponds and crops and/or pesticide use which exist in real agricultural landscapes. Standard regulatory assumptions used to estimate spray drift and runoff were modified in order to estimate distributions of exposure for 597 ponds in an intensive cotton-growing county in Mississippi. Modifications were made on the basis of the following data:

- Landscape data on the proximity of cotton to ponds and the amount of cotton in the watershed, derived from Remote Sensing and GIS (Geographic Information System).
- The no-spray buffers that are mandated on pyrethroid cotton labels in the US between aerial and ground applications and surface waters.
- The percentage of cotton acres treated with pyrethroids.

Inclusion of these factors significantly reduced the predicted exposure concentrations compared to the standard regulatory assumptions, and allowed the development of more environmentally realistic probabilistic exposure distributions for Mississippi ponds. In addition to the small number of factors included in the analysis, a range of further landscape factors (e.g. effects of buffer vegetation and wind direction on spray drift) would be expected to further reduce the amount and/or the frequency of exposure in reality.

PWA152 Probabilistic Aquatic Risk Assessment of Pyrethroids: 5. Combining Effects and Exposure to Characterize Risk. Maund*, S.J., Travis, K.Z., Zeneca Agrochemicals, Bracknell, UK; Hendley, P. Zeneca Ag Products, Richmond, CA; Solomon, K.R., University of Guelph, Guelph, ON; Giddings, J.M. Springborn Laboratories, Inc., Wareham, MA. On behalf of the Pyrethroid Working Group (PWG). Here we describe the application of probabilistic approaches to risk characterisation for uses of the cypermethrin in US cotton. Our initial application of the probabilistic approach was to compare estimated exposure concentrations derived from GENEEC (Tier I) and PRZM/EXAMS (Tier II) models to a value derived from the probabilistic effects distribution. Previous studies have suggested that the 10th centile from the effects distribution is probably a suitable value and will be conservative of effects under field conditions. For cypermethrin, Tier I and II assessment identified a marginal level of concern for fish, and a clear level of concern for water-column and sediment invertebrates. However, modified Tier II probabilistic exposure concentrations, which incorporated landscape-level data to refine the model assumptions, indicated no concerns for fish or sediment invertebrates, and only a marginal level of concern for water-column invertebrates. Comparison of these exposure concentrations to effect concentrations from field studies also indicated a large margin of safety. As a more sophisticated application, we developed probabilistic distributions of risk quotients. The degree of overlap between the exposure and effect distributions was quantified by comparing each exposure value with each effect value from the respective distributions, which were then placed in a ranked distribution. For cypermethrin, this approach demonstrated that there was little likelihood of exposure concentrations occurring which would lead to adverse effects under agriculturally realistic conditions.

PWA153 An Probabilistic Ecological Risk Assessment Method Evaluating Cumulative Episodic Exposure to Chemical Stressors Applied to a South Carolina Tidal Stream Community Impacted by Agricultural Runoff. M. G. Morton, U.S. Environmental Protection Agency Region 6, 1445 Ross Avenue, Dallas, TX, F. L. Mayer, Gulf Ecology Division (ORD/NHEERL) U.S. Environmental Protection Agency, Sabine Island, Gulf Breeze, FL, K. L. Dickson, W. T. Waller, M. F. Acevedo, University of North Texas, Institute of Applied Sciences, P.O. Box 13078, Denton, TX . A prospective ecological risk assessment method was developed evaluating the cumulative probabilistic impact of chemical stressors to aquatic organisms. A probabilistic expression of the percent of an ecosystem's species at risk is generated from a designated chemical exposure scenario. This method, which evaluates the magnitude, duration and episodic nature of chemical stressors on communities, was used to predict the ecological risk associated with agricultural runoff to an estuarine system. *In situ* data collected from a multi-year agricultural pesticide runoff study conducted in Leadenwah Creek, South Carolina were used for field verification of the method. For effects assessment, probabilistic distribution (extrapolation) methods use laboratory toxicity tests to generate stressor distribution curves for the pesticides. Stressor estimated safe concentrations (ESC) for the estuarine ecosystem are selected from the distribution curve at $P = 0.05$. For exposure assessment, fate and transport modeling was employed to generate temporal stressor exposure concentration curves in Leadenwah Creek for azinphos-methyl and fenvalerate from sparse measured data. An "Area Under the Curve" (AUC) integration is performed on predicted exposure concentrations exceeding the ecosystem's stressor ESCs to calculate cumulative adverse pesticide exposure over time. The integrated stressor exposure concentrations are adjusted for an allowable duration of exposure to the stressor ESC. The stressor cumulative exposure concentration is then applied to the stressor's distribution model (curve) to predict the percent of the ecosystem's species at risk to the cumulative magnitude and duration of an episodic exposure event. Additivity was assumed for simultaneous exposure to multiple pesticides. Limited biological data were available for validation of the prospective method. However, the probabilistic ecological risk assessment predictions compared well with the Leadenwah Creek *in situ* bioassay results. With modifications, the method may be used to predict ecological risk from chemical exposure to other communities.

PWA154 Probabilistic Aquatic Risk Assessment of Cotton Pyrethroids. Maund, S.J.*, Travis, K.Z., Zeneca Agrochemicals, Bracknell, UK; Giddings, J.M., Springborn Laboratories, Wareham MA; Solomon, K.R., University of Guelph, Guelph, ON; Hendley, P., Zeneca Ag Products, Richmond, CA. On behalf of the Pyrethroid Working Group (PWG). Current international approaches to regulatory risk assessment for pesticides typically utilise single estimates of effects and exposure concentrations to characterise potential risks. More recently, interest in the application of probabilistic approaches to risk assessment has been developing. One advantage of using a probabilistic approach is that it gives a better indication of the range of risks by utilising the entire toxicity and exposure datasets, rather than relying on a single effect and exposure concentration. Here we summarize a series of studies which were performed to develop a probabilistic risk characterisation for cotton uses of pyrethroid insecticides in the USA. Distributional analyses of laboratory toxicity data were used to describe effect concentrations for fish and aquatic arthropods. These were calibrated with extensive data from the large database of field studies available for pyrethroids. Probabilistic exposure concentrations were developed by modifying standard models (PRZM/EXAMS) with landscape-level data (satellite and aerial images) on the co-occurrence of cotton and ponds in Yazoo County, Mississippi, USA (a 'worst-case' cotton county). Probabilistic risk characterisation was performed by developing a distribution of risk quotients that combined the probabilistic exposure and effect distributions. The assessment showed that it was unlikely that pyrethroids would have adverse impacts on ponds in Yazoo County, principally because the majority of cotton fields are relatively distant from water bodies.

PWA155 Application of Approaches for Evaluating the Bioavailability of Chemicals in Terrestrial and Aquatic Environments L.A. Baron*, McLaren/Hart, Inc.-ChemRisk, Warren, NJ.; N.L. Bonnevie, McLaren/Hart, Inc.-ChemRisk, Portland, ME; C.R. Harman, P.W.S., McLaren/Hart, Inc.-ChemRisk, Warren, NJ. In both terrestrial and aquatic risk assessments, it is important to consider site-specific conditions that affect the bioavailability and ultimate toxicity of chemicals in the environment. Consideration of such conditions provides a more realistic estimate of actual potential risks to ecological receptors. For example, recent developments regarding the bioavailability of metals in sediments have demonstrated that many metal ions form sulfides and organic complexes that make them non- or less bioavailable to aquatic organisms. Similar binding mechanisms influenced by pH, cation exchange capacity (CEC), organic carbon, and oxidation reduction potential (Eh) affect the bioavailability of metals in surface soils to terrestrial receptors (e.g., earthworms, plants, and vermivorous/herbivorous wildlife). Specifically, evidence of decreased bioavailability was observed through a reduction of metal uptake by plants and earthworms in the terrestrial environment. The actual exposure of ecological receptors to chemicals is a primary factor which regulates bioaccumulation and toxicity, particularly for higher trophic-level species. In this evaluation, we present the results from several case studies demonstrating the importance of evaluating potential bioavailability of sediment- and soil- associated contaminants. Approaches for evaluating the bioavailability of metals will be discussed, as well as their application to ecological assessments.

PWA156 The Impact of Dissolved Organic Matter on the Bioavailability and Uptake of Hydrophobic Xenobiotics in Juvenile Rainbow Trout. P. Qiao and A.P. Farrell, Departments of Biological Sciences, F.A.P.C. Gobas, School of Resource and Environmental Management, Simon Fraser University, Burnaby, BC. This study measured tissue concentrations of 1,2,4-trichlorobenzene (1,2,4-TCB), 1,2,3,4,5-pentachlorobenzene (PeCB) and 2,2',4,4',6,6'-hexachlorobiphenyl (HCBP) (log Kow values from 3.95 to 7.55) in juvenile ra in! fish exposed to 1.54 mg/L humic acid compared with controls. Thus humic acid concentration in the range of 1-5 mg/L reduced the bioavailable chemical concentration for a chemical with a very high log Kow value, e.g., 7.55. However, evidence is provided to indicate that humic acid concentrations between 1-5 mg/L may result in increased bioaccumulation of 1,2,4-TCB and PeCB.

PWA157 Factors influencing mercury concentrations in yellow perch from Kejimkujik National Park, Nova Scotia. Burgess, N.M.*, Canadian Wildlife Service, Sackville, NB; d'Entremont, A., d'Entremont Environmental Solutions, Hammonds Plains, NS; Drysdale, C., Parks Canada, Kejimkujik National Park, NS; Vaidya, O., Environment Canada, Dartmouth, NS; Brun, G.L., Environment Canada, Moncton, NB. As part of a larger study on the bioaccumulation and ecological effects of mercury in freshwater fish and wildlife in Atlantic Canada, we measured total mercury in yellow perch (*Perca flavescens*) from 24 lakes in Kejimkujik National Park, Nova Scotia. Objectives of the study were to assess mercury concentrations in prey of common loons (*Gavia immer*), and determine which environmental factors were associated with differences in fish mercury levels between lakes. From each lake, we tried to collect 27 perch, 9 from each of three size-classes (5-10, 10-15 and 15-20 cm fork length). A total of 678 yellow perch were collected and 242 composite samples of (usually) three whole fish each (grouped according to length) were analysed for total mercury. Mercury concentrations ranged from 0.05 to 0.77 ug/g in composite samples with average fish weights of 1.9 to 105 g and ages of 1 to 10 years. Mercury concentrations were positively correlated with fish length, weight and age ($r_s > 0.7$). Significant differences were found between lakes in rates of mercury accumulation with fish size and in the size-adjusted mean perch mercury concentrations. These differences in mean fish mercury concentrations correlated negatively with lakewater pH and positively with aluminium concentrations ($r_s = 0.6$), but not with total mercury concentrations in lakewater. Multivariate analysis was used to assess which lake chemistry parameters and watershed characteristics were most predictive of yellow perch mercury levels. Methylmercury concentrations and stable carbon and nitrogen isotope ratios were also measured in a subset of the fish samples.

PWA158 Effects of Temperature on the Uptake of Benzo[a]pyrene by Isolated Hepatocytes of Rainbow Trout, *Oncorhynchus mykiss*. Johnston, B.D.* and Kennedy, C.J., Simon Fraser University, Burnaby, BC, Canada. Aquatic ectotherms, such as teleosts, are adapted to their thermal environments by a host of physiological and biochemical mechanisms which may confer differences in the toxicokinetics of xenobiotics. Thermally-induced changes in toxicokinetics of benzo[a]pyrene (BaP) have already been demonstrated in uptake by gill cells, and in biotransformation rates by liver cells. However little information is available regarding the mechanisms of uptake or excretion of xenobiotics by teleost hepatocytes. The objectives of this study were therefore to determine the method of uptake of BaP by rainbow trout hepatocytes, and to determine how temperature effects this process. Liver cells were isolated from animals acclimated to 8û or 18û C and exposed to BaP in a modified HankÕs salts solution at several temperatures. There were no significant differences in uptake rates between hepatocytes acclimated and exposed at the same temperature. However, hepatocytes acclimated to 18û C and exposed to BaP at 8û C showed a 2-fold decline in uptake rates. Preliminary experiments indicate that incubation with ouabin has no effect on uptake rates of BaP. These results indicate that acute temperature change alters the rate of uptake of benzo[a]pyrene by rainbow trout hepatocytes. However, a high degree of temperature compensation in hepatocyte uptake rates is preserved over acclimatory time-spans. Preliminary Na^+ transport manipulations indicate that uptake of BaP into hepatocytes is by passive diffusion.

PWA159 Water Quality Assessment of The Pirajussara River (São Paulo-sp, Brazil): First Results. Abessa, D.M.S., University of São Paulo, São Paulo, SP, Brazil. In the recent years, the conservation, managment and remediation of the water resources have claimed for more attention by the publics and the researchers. This study had the purpose to evaluate preliminarly the quality of the waters of Pirajussara River, by acute toxicity testing and physical-chemical analysis. Four campaigns were made, in each one samples from 4 sites were collected. Low D.O. concentrations and high ammonia concentrations were observed in all the samples. Acute toxicity on *Daphnia similis* were observed in all the samples of the first two tests, in two samples of the third and the fourth tests. The results showed that the water of the Pirajussara River do not present suitable quality for the aquatic life, probably due to the sewage pollution. Further studies containing chemical analysis must be run aiming to identify the compounds responsible by the toxicity.

PWA160 Determination of Conditional Chemical Equilibrium Constants for Binding of Metals to a Common Invertebrate. Wade, P.W.* and Todd, C.P., CSIR, South Africa. *Daphnia pulex* were exposed to metal ions, including Cu^{2+} and Zn^{2+} in acute mortality tests. In all tests, Ca^{2+} was present, to quantify the antagonism of hardness on metal toxicity. The resulting fractional mortality vs concentration data was analysed by Logit transform, combined with some thermochemical equations, to yield the conditional metal binding constants and the concentrations of receptor sites on the invertebrates.

PWA161 Repetitive Check Sample Results for PCBs and Trans Nonachlor in Lake Trout (*Salvelinus namaycush*). Chernyak, S.M.*, Begnoche, L.J., U.S. Geological Survey, Ann Arbor, MI; Rice, C.P., U.S. Department of Agriculture, Beltsville, MD; Quintal, R.T., Hesselberg, R.J., Schmidt, L.J., Hickey, J.P., U.S. Geological Survey, Ann Arbor, MI. Eighty-one replicate analyses of the Great Lakes Science Center Check Fish (1989 Lake Ontario Lake Trout, five 4-year olds 4 male and 1 female, 600-700 mm in length and weighing 2-3 kg, stored at -20 degrees C) were carried out from June 5, 1996 till March 6, 1997. The results for these analyses include several PCB congeners (ten as coeluting pairs, five as separately isolated peaks) and one pesticide byproduct (trans nonachlor). Lipid contents of each separate extraction were also measured. This check sample has been used off and on over several years to validate consistency of analytical performance at the laboratory and to provide a benchmark for comparison into the future. The coefficient of variation (CV) for the organochlorine residues varied from a low of 6.63 for congener 174 to the highest value of 19.62 for the combined congener peaks for congener 31+28. The average relative percent differences were consistently lower than the CV's by at least 16%. The analyte concentrations varied very little over the time coarse the experiment whether correlated to extraction time of subsequent analyses date. Results for recovered lipid versus recovered analyte amounts will be considered. The data will also be examined for any associative trends between or among any of the separate analytes.

PWA162 Polycyclic Aromatic Hydrocarbon Accumulation in a Trophically Important Benthic Copepod Living in Two Estuarine Creeks. Klosterhaus, S.L.*, University of South Carolina, Columbia, SC; Ferguson, P.L., SUNY-Stony Brook, Stony Brook, NY; Chandler, G.T., University of South Carolina, Columbia, SC. Bioaccumulation of polycyclic aromatic hydrocarbons (PAHs) by benthic organisms living in contaminated sediments may vary considerably by site, depending on contaminant concentration and sediment characteristics. To gain insight into factors affecting PAH bioaccumulation, we measured concentrations of three PAHs in the benthic estuarine copepod, *Microarthridion littorale* and in its surrounding sediment collected from two contaminated sites on the coast of South Carolina, USA. Both sites harbor abundant natural populations of *M. littorale*, which is a primary food of juvenile fish and shrimp. Diesel Creek is an EPA Superfund site located near Charleston Harbor, Charleston, SC. Marina Pipe is a moderately urbanized estuarine creeklet located adjacent to a residential community in Murrell's Inlet, SC. Concentrations of the PAHs Fluoranthene (FLU), Benz[a]anthracene(BAA), and Benzo[a]pyrene (BAP) were determined in sediments and in *M. littorale* tissues using GC-MS with stable isotope dilution quantification. FLU tissue concentrations were higher and BAA and BAP tissue concentrations were lower than sediment concentrations at both sites. Assuming 10% lipid content in *M. littorale*, BSAFs at Marina Pipe were 0.463 for FLU, 0.212 for BAA, and non-quantifiable for BAP since tissue concentrations were undetectable. *M. littorale* BSAFs at Diesel Creek were 0.654 for FLU and 0.130 for BAA. Concentration dependent differences found for FLU bioaccumulation but not for BAA may be due to an inducible metabolic pathway for compounds with high log K_{ow} (i.e. BAA) in *M. littorale*. These BSAFs represent pooled collections of adult males, and gravid/non-gravid females. Present analytical efforts are determining whether sex and reproductive condition affect copepod lipid states, uptake rates, and BSAFs.

PWA163 Environmental Chemistry and Mercury Uptake by Algae. Petri, B.*, York University, North York, Ontario; Mierle, G., Ontario Ministry of Environment and Energy, Dorset, Ontario; McQueen, D., York University, North York, Ontario. The largest bioconcentration factor for mercury in aquatic ecosystems occurs between the water and the seston. Water chemistry determines mercury and methylmercury speciation, and can thus affect uptake rates into algae and overall bioaccumulation in higher trophic levels. We measured inorganic mercury and methylmercury uptake rates into algae (UTEX 20) using defined media to determine transport complexes and membrane permeabilities. Using natural waters, the effects of organic complexing agents on mercury bioavailability were evaluated. Speciation of Hg(II) and $CH_3Hg(I)$ was calculated using MINEQL. Uptake experiments were performed under clean room conditions. Aliquots were filtered over a set time course, and uptake rates were determined from the initial slopes. We determined that both neutral chloride and neutral hydroxide species were taken up by algae. For either Hg(II) or $CH_3Hg(I)$, chloride and hydroxide permeabilities were of the same magnitude, but permeabilities of $CH_3Hg(I)$ complexes were 4 to 5 fold higher than for Hg(II) complexes. Organic complexing agents reduced uptake rates and thus bioavailability.

PWA164 Use of Passive Sampling Devices to Detect Organochlorine Contaminants in Morelet's Crocodile *(Crocodylus moreletii)* Habitat in Belize. Rainwater, T.R.,* Anderson, T.A., McMurry, S.T., The Institute of Environmental and Human Health, Texas Tech University, Lubbock, TX ; Johnson, K.A., Department of Chemistry, Southern Illinois University at Edwardsville, Edwardsville, IL; Platt, S.G., Lamanai Field Research Center, Indian Church Village, Orange Walk District, Belize. Recent studies have demonstrated the sensitivity of crocodilians to environmental contaminants, particularly organochlorines (OCs). Due largely to their high trophic status, small home range, and long life span, crocodilians readily bioaccumulate OCs, and exposure to these compounds may result in numerous adverse reproductive effects. In Central America, regulations governing the production, distribution, and use of pesticides are scant or inadequately enforced. Therefore, crocodilians and other wildlife inhabiting these areas may have an increased risk of chemical exposure. We are currently conducting a study to examine exposure and resulting impacts of OCs on the endangered Morelet's crocodile *(Crocodylus moreletii)* in Belize. One primary objective is to characterize OC contamination of crocodile habitat within the study area. In June 1997, we deployed passive sampling devices (PSDs) along the banks of the New River watershed and Gold Button Lagoon, two systems in northern Belize known to support relatively large populations of Morelet's crocodiles. PSDs are highly efficient tools in contaminant assessment as they increase the ease and speed of analysis, decrease solvent usage and cost, minimize the transport of contaminated material, and improve detection limits through sample concentration. PSDs remained in the field for approximately two months, at which time they were collected and stored at approximately -25 °C until analysis. Each PSD was screened for 20 OCs using GC/ECD. Based on retention times compared to authentic standards, *p,p*'-DDD and heptachlor (among others) appear to be present in both watersheds. The source(s) of these contaminants is unknown, although past or current use of OC pesticides in nearby agricultural areas is possible.

PWA165 Selective Retention of Toxaphene in Saltmarsh Sediments and Biota. Maruya, K.A.*, Wakeham, S.G., Skidaway Institute of Oceanography, Savannah, GA; Francendese, L., U.S. EPA, Atlanta, GA. As part of an assessment of the ecotoxicological risk associated with toxaphene, sediments and biota were collected from several locations within a tidal saltmarsh adjacent to a former toxaphene manufacturing plant in Brunswick, Georgia (USA). These samples were analyzed by GC-ECD, GC-electron impact MS (EIMS), and GC-negative chemical ionization MS (NCIMS) using 30m glass capillary columns. Levels of toxaphene components, or polychlorinated camphenes (PCCs), were high enough to be routinely detected by GC-EIMS. Compared with technical toxaphene (supplied by the toxaphene manufacturer), the PCC profile decreased in complexity from sediments to resident fish to migratory forage fish to predator fish. Moreover, the predominant peaks (Cl_6-Cl_8 homologs) in sediment and tissue samples were shifted toward earlier eluting PCCs. Using GC-NCIMS, the most prominent peak in all samples was a hexachlorinated PCC eluting early in the technical toxaphene chromatogram. These results indicate that (i) reductive dechlorination of PCCs may be occurring in these sediments; (ii) resident fish retain the PCC profile associated with sediments collected from the same location; and (iii) fish at higher trophic levels selectively accumulate certain PCCs.

PWA166 Metal-Colloid Partitioning in Interstitial Waters of Marine Sediments: Influences of Salinity, pH and Colloidal Organic Carbon Concentration. Cantwell, M.G.*, Burgess, R.M., U.S. EPA, Narragansett, RI. For decades, heavy metals have been deposited into marine sediments as a result of anthropogenic activities. Depending on their bioavailability, these metals may represent a risk to benthic organisms. Dissolved metal concentrations have been shown to be better predictors of bioavailability than the total sediment metal. In order to better predict dissolved concentrations, it is essential to understand metal speciation. In sediment, interstitial water is a primary route of metal exposure to many benthic organisms. Colloidal organic carbon is ubiquitous in marine interstitial water and readily forms complexes with numerous dissolved metals greatly reducing their bioavailability. We are currently developing a methodology for isolating dissolved and colloidal cadmium, copper, nickel, lead and zinc from interstitial water in order to derive partition coefficients. Interstitial waters are separated from sediments by centrifugation followed by colloidal and dissolved metal isolation by stirred cell ultrafiltration. Three types of samples are being evaluated: metal-spiked humic acid solution, metal-spiked reference sediment, and environmentally contaminated sediment. Variability in humic acid partition coefficients (Kp) as a function of colloidal organic carbon concentration were greatly reduced by deriving the colloidal organic carbon partition coefficient (K_{coc}). Based on K_{coc}, cadmium (3.99) had the lowest affinity for colloidal organic carbon while copper (6.36) had the highest. As expected, K_{coc} for the metals increased as pH increased and salinity decreased. Development of partition coefficients for individual toxic metals normalized for colloidal carbon will improve our ability to predict metal bioavailability in marine sediments.

PWA167 A Combined Desorption/bioaccumulation Study on PAH's in Aged and Non-aged Contaminated Sediments. Kraaij, H.*, Research Institute of Toxicology, Utrecht; Cornelissen, G., Institute for Inland Water Management and Waste Water Treatment, Lelystad; Belfroid, A., Institute for Environmental Studies, Free University, Amsterdam; Tolls, J., Research Institute of Toxicology, Utrecht, The Netherlands. Unpolluted sediment was spiked with 7 PAH's with a MW 178 to 252. Directly after spiking, oligochaetes (tubificidae) were exposed to the contaminated sediment for a maximum of 35 days and analyzed using a MSPD (Matrix Solid Phase Dispersion) technique. The experiment was repeated with the contaminated sediment that had been stored for three months at 10 °C. After 0, 0.5 and three months after spiking, desorption kinetics were determined on subsamples of the contaminated sediment being used in the accumulation experiments. The desorption curve was obtained using a tenax extraction technique (Cornelissen et al, 1997). Results of the bioaccumulation experiment and the desorption experiment for the non-aged and three months stored spiked sediment were compared to see if three months of aging had an effect on bioavailability and desorption kinetics. Both accumulation in tubificidae and desorption kinetics did not change for the aged contaminated sediment compared to the non-aged contaminated sediment for all PAH's. BSAF's (biota to sediment accumulation factor) decreased significantly with increasing MW. The percentage of the PAH's in a slowly desorbing fraction (k of desorption = ca. 5EE-3 h) was constant at 15-45 % of the total fraction of PAH's in the sediment. Cornelissen G et al 1997 Desorption kinetics of chlorobenzenes, polycyclic aromatic hydrocarbons, and polychlorinated biphenyls: sediment extraction with tenax® and effects of contact time and solute hydrophobicity. Environ. Toxicol. Chem. 16 (7), 1351-1357.

PWA168 Influence of Sulfide on the Partitioning of Cd, Zn and Ni in the Porewater and Sediments. Yi, J.-S.*, Koh, C.-H., *Seoul Nat'l Univ. S.Korea*; Lee, B.-G., Jeon, H.-S., Luoma, S.N., *USGS, Menlo Park, CA.* Laboratory microcosm experiments were conducted to investigate the influence of AVS (acid volatile sulfide) on the partitioning of Cd, Ni and Zn in pore-water and SEM (simultaneously extracted metal). Eight experimental sediments were prepared by spiking the same amounts of Cd, Ni and Zn (5, 280, 390 μg/g, respectively) into sediments containing four levels of AVS (0.5, 6, 15, 35μmole/g) or by adding four levels of Cd (2~17μg/g), Ni (55~620μg/g) and Zn (110~830μg/g) to sediments having a constant AVS (6μmole/g). Following 18d incubation at 15°C, AVS in the surface sediments (<0.5cm) decreased by 65~95% due to oxidation, while initial AVS was maintained at depth. The AVS in the sediments treated with different levels of metals increased with SEM, suggesting that the spiked metals retarded sulfide oxidation. During the incubation period all three metals maintained their initial SEM levels with little vertical gradient. Nearly 100% of the spiked Cd and Zn was recovered as SEM, but only 50~60% of the Ni was recovered. The levels of Cu extracted as SEM were highest at the oxidized surface or in the sediment with lower AVS. Pore-water Cd levels were affected considerably by AVS; the highest pore-water Cd occurred in the surface sediments with low AVS. The pore-water Ni and Zn increased linearly with the molar difference of (SEM-AVS) and increased rapidly when the difference was > 0. The partition coefficient (k_d), estimated from the pore-water metals and (SEM-AVS), was 1200 for Zn and 70 for Ni. Diffusion into overlying waters was evident when high pore-water Ni and Zn were present in the surface.

PWA169 Concentrations and Partitioning of PAH's in Danish Marine Sediments. Pritzl, G.*, Nat. Env. Res. Inst., Roskilde, Denmark; Poulsen, M. E., Nat. Vet. & Food Admin., Soeborg, Denmark; Larsen, H. County of S. Jutland, Toender, Denmark; Foverskov, S., Nat. Env. Res. Inst., Roskilde, Denmark. In a study with special regard on the regulation of dumping of dredged harbour sediments, a survey of twentytwo PAH's and dibenzothiophene's in Danish marine estuarian sediments from stations ranging from contaminated harbours to open sea locations evinced concentrations (as sum of PAH's) between 150 ng/g d.w. and 8000 ng/g d.w. The chronic toxicity (measured as benzo(a)pyrene equivalents) and the acute toxicity (measured as 2-methyl naphthalene equivalents) were a factor of 10 to 50 times lower. The salinity ranged between 0.9 % to 3.2 % and the content of organic carbon from 0.9 % to 12 %. These factors implicated on the bioavailability of the different PAH's, measured as the truly dissolved concentrations, following a compartmentalization of the porewater by centrifugation at 20000g and reverse-phase separation by a C-18 column.

PWA170 A Comparison of the Relative Contributions of Gill and Gastrointestinal Tract to the Uptake of Three Hydrophobic Chemicals. P. Qiao and A.P. Farrell, Departments of Biological Sciences, F.A.P.C. Gobas, School of Resource and Environmental Management, Simon Fraser University, Burnaby, BC. This study was designed to assess the relative contributions of gill and GI tract uptake for 1,2,4-trichlorobenzene (1,2,4-TCB), 1,2,3,4,5-pentachlorobenzene (PeCB) and 2,2',4,4',6,6'-hexachlorobiphenyl (k! d values, a simulation model was used to predict the relative contributions of gill and GI tract uptake when fish were exposed simultaneously to contaminated water and food. The model predicted that the two uptake routes would contribute equally to the chemical body burden in fish when the food:water chemical concentration ratio was near to 105 for these three test chemicals. According to the expected food:water chemical concentration ratios in nature, gill uptake was predicted to account for over 98%.

PWA171 Pesticide Bioavailability Using an *In vitro* Test. Davis, A., Geomega, Boulder, CO; Tabor, C., Geomega, Boulder, CO, Scofield, R., Environ, Emeryville, CA, Meyer, M.C.*, Geomega, Boulder, CO. An *in vitro* test that assesses the bioaccessability of pesticides has been developed and tested on soils containing chlordane, DDT, and DDE. The method uses a system that mimics the conditions of the stomach and intestinal phases of the mammalian gastric system, with the goal of measuring the fraction of the pesticides in soil that are solubilized by the gastric solutions. Bioaccessability, or the mass percent of pesticides solubilized versus the mass in the soil, is used as a surrogate measure of bioavailability for the different chlorinated pesticides. Stomach conditions are mimicked by a solution of hydrochloric acid, pepsin, citrate, malate, lactic acid, and acetic acid at pH 2.5 for 1 hour. Sodium bicarbonate is then used to raise the pH to 7.0 (~1 hour), upon which time, bile salts and pancreatin are added. An additional 3 hour incubation follows. Pesticide solubility is assessed at several time points during both the stomach and small intestine phases of the experiment. This method resulted in an average 3.1% bioavailability for chlordane, and <4.2% and <5.6% bioavailability respectively for DDT and DDE.

PWA172 Detection and Quantification of Nitric Oxide Desorbed From Ambient Air Particles And Particle Emissions From Vehicles. Ball, J.C.*, Hurley, M.D., Straccia, A.M., Gierczak; C.M., Ford Motor Co., Scientific Research Laboratory, Dearborn, MI. In light of the numerous reports of an association between particle air pollution and adverse human health effects, it is important to chemically characterize ambient and source-related particles. Among the gases which might be adsorbed onto such particles, nitric oxide is of interest because it naturally occurs in the human body and has physiological effects such as vasodilation. In this work, NO desorbed from four NIST reference materials, two ambient air particulate samples and two diesel particulate samples, is identified and quantified after heating the samples for one hour. Observed NO concentrations ranged from 0.005 ng NO / mg sample at 37 °C to 1900 ng NO / mg sample at 140 °C. In addition, NO desorption was measured from particles collected from six late model spark-ignition vehicles after heating the particles at 120 °C for one hour. NO concentrations ranged from 50 ng NO / mg to 560 ng NO / mg particulate sample. Measurements of NO desorption at 37 °C from particles collected from spark-ignition vehicles were not feasible because the particle mass emission rates for current technology vehicles are too low, around 1-2 mg/mile, to permit collection of adequate amounts of particulate material.

PWA173 Observations on Declining SPMD Sampling Rates for High K_{ow} Compounds. Huckins, J.N.*[1]; Petty, J.D.[1]; Gale, RW.[1]; Booij, K.[2]; Prest, H.F.[3]; Clark, R.C.[1]. USGS, Columbia, MO.[1]; Netherlands Institute for Sea Research, The Netherlands [2]; and Long Marine Laboratory, Santa Cruz, CA [3]. McKim and other investigators have observed a significant decline in the uptake fluxes of chemicals by fishes, as log K_{ow}s exceed ≈ 7. Often this phenomenon occurs at water concentrations well below chemical saturation; thus water solubility does not seem to be the limiting factor. A similar phenomenon has been reported for lipid containing Semipermeable Membrane Devices (SPMDs), which in theory sample only dissolved phase chemicals. In this work, we examine both SPMD and biota data and explore a number of factors that may play a significant role in the decreasing-flux phenomenon. These factors include water-borne sorbates, boundary layer and membrane resistances, biofouling, and lipid solubility. Mass transfer theory related to aqueous boundary layers does not appear to justify the steepness of the slope of declining uptake rates for increasingly hydrophobic compounds. An implicit absorbate can be evoked to adjust sampling rates to those predicated by theory, but the validity of this approach is not certain. For SPMDs, biofouling may account for increased impedance to mass transfer with increasing K_{ow}, but this is clearly not the case for biota. These and other factors potentially contributing to decreasing sampling rates will be examined in some detail.

PWA174 Influence of Citrate on Accumulation of Cadmium by Durum Wheat. Berkelaar, E. J.* and Hale, B. A., University of Guelph, Guelph, ON, Canada. Metal ions which are dissolved in solution exist as the free ion (M^{z+}) or one of several metal-ligand complexes ($M\text{-}L_n^{\circ}$), depending on the solution. The results of experiments measuring the effects of dissolved metals on various aquatic organisms have suggested that the free ion is the bioavailable form of the metal. This has led to the formulation of the Free Ion Model (FIM) to explain the effects of metals dissolved in solution on organisms. More recently, however, exceptions to the FIM have been noted. In studies with aquatic organisms, the dissolved metal was more bioavailable when in the presence of certain aqueous ligands (L). For this study, several experiments were carried out with the objective of determining the relationship between accumulation of Cd in plant roots and the presence of citrate in the exposure solution. Our null hypothesis was: accumulation of Cd by roots of durum wheat (*Triticum turgidum*) is dependant only on the concentration of free Cd ion (Cd^{2+}), and is not influenced by the presence of citrate. Hydroponically grown durum wheat seedlings were transferred to a simplified nutrient solution containing Cd^{2+}, either with or without citrate. Root Cd concentration as well as total Cd and Cd^{2+} in the exposure solutions were determined. The effects of citrate on other aspects of solution chemistry, such as Ca^{2+} and Mg^{2+} were also monitored. Our results show that at similar Cd^{2+} concentrations, the presence of citrate resulted in greater accumulation of Cd than predicted. In solution, citrate complexes with Cd^{2+}, Ca^{2+} and Mg^{2+}. Enhanced accumulation of Cd may be due to accumulation of Cd-citrate complexes, or a reduction in competition for uptake sites by Ca^{2+} and Mg^{2+}. Therefore, we conclude that in this case, accumulation of Cd by roots of durum wheat was not only dependent on the concentration of Cd^{2+}, but was enhanced by the presence of citrate.

PWA175 The Effect of pH on Bioavailability and Partitioning of Organic Contaminants in Sediments. Standley, L. J.*, Stroud Water Research Center, Academy of Natural Sciences, Avondale, PA. In previous research on bioavailability and partitioning of contaminants in natural sediments, bioaccumulation of dieldrin by the aquatic earthworm *Lumbriculus variegatus* was strongly, but inversely correlated with pH ($r^2 = 0.899$, $p < 0.05$). However, because sorption of contaminants by organic matter is generally reduced with increasing pH and sorption is believed to reduce the bioavailability of contaminants, this result was counterintuitive. Thus, experiments were conducted to confirm the relationship between bioavailability of nonpolar organic contaminants and sedimentary pH. Sediments were spiked with γ-hexachlorocyclohexane (lindane), 2,2',5,5'-tetrachlorobiphenyl (PCB52), dieldrin, 2,2',3,3',5,5'-hexachlorobiphenyl (PCB133), and benzo(a)pyrene in a methanol carrier, treated with phosphate buffer to adjust pH, and stored at 4ûC for 8 days to equilibrate. *L. variegatus* were exposed to amended sediments for 3 d at 15ûC. Additional sediment samples for analysis of contaminant partitioning behavior were treated in the same manner. Pore waters were isolated from sediments by centrifugation and filtration. Water samples were split and half extracted by liquid-liquid extraction (total contaminant) and half extracted following elution through C-18 sep-paks (DOM-bound residues). Freely-dissolved residues were determined by subtraction. Extracts were analyzed using GC/MS (selected ion monitoring).Results for pH-adjusted sediments confirmed the inverse correlation measured previously between bioavailability and pH in unamended sediments. Analysis of freely-dissolved residues in pore waters confirmed the expected trend of increasing concentrations with increasing pH. Thus, bioavailability of contaminants to *L. variegatus* were not explained by partitioning of contaminants between sedimentary compartments as determined using solvent extractive techniques.

PWA176 Influence of Dissolved Organic Matter on UV Photolysis of PAHs. Tarr, M.A.*; Floyd, W.T., Jr., University of New Orleans, New Orleans, LA. Polycyclic aromatic hydrocarbons (PAHs) are known to undergo direct photolysis in sunlight. Generally, ultraviolet (UV) radiation is responsible for these phototransformations. Although significant work has been done on atmospheric and solid phase photoreactions of PAHs, little work has been done in aqueous environmental systems. The presence of dissolved organic matter (DOM) in natural waters can have significant impacts on PAH phototransformations. The effects of DOM may include: 1) protection of PAHs by light screening effects, 2) enhancement of degradation through photosensitization, and 3) alteration of PAH direct photolysis rates through PAH-DOM binding. We have observed first order degradation of pyrene in pure water and 30 mg/L Suwannee River fulvic acid. In both cases the same single fluorescent product was observed. The photoproduct also degraded at a similar rate as the pyrene. No other products were observed by fluorescence, although non-fluorescent products may be present. Pyrene in the fulvic acid sample showed more rapid degradation than in pure water, indicating that DOM sensitized photolysis predominated in the fulvic acid solution. This result indicates that previously measured direct photolysis rates are likely to overestimate PAH photolysis half lives in natural waters. Furthermore, the presence of DOM results in enhanced PAH water solubility, thereby increasing the aqueous load of PAHs. Together, these effects may have significant impacts on the lifetime of PAHs in aquatic environments. Both the photoproduction and photodegradation rates of detrimental photolysis products (toxic, carcinogenic, endocrine disruptive) are likely altered by DOM.

PWA177 Characterization of Dissolved Organic Matter Using Fluorescence. Smith, D.S., McMaster University, Hamilton, ON, Canada; Kramer, J.R., McMaster University, Hamilton,ON, Canada. Dissolved organic matter (DOM) samples from nine different sites in Norway are characterized with regards to numbers and types of fluorophores. Contributions of individual fluorophores are extracted from measured exciation versus emission fluorescence sufaces using spectral deconvolution. These fluorophores are interpretted relative to the fluorescence of model compounds. In general, the samples require between four and seven components to describe the measured fluorescence. Proposed fluorescent moities include: simple aromatics, flavones, coumarines and quinoid compounds. Definitive identification of fluorophores is not possible because there is significant overlap between the groups of compounds. These structural units may occur as free molecules or as building blocks of macromolecules. Initially, the samples contained aluminum concentrations in the 1.4 to 23 μmol/L range. Addition of extra aluminum to the samples caused only minor changes in position of components, but fluorescence intensites increased. This is interpretted as evidence for substantial contributions of aluminum-bound fluorophores in DOM fluorescence. It was found that younger organic matter from lakes with shorter residence times required fewer components, but older DOM was more complicated and more components were required to explain the fluorescence observations.

PWA178 Carbonate Radical Reaction Pathways towards Sulfur-containing Aromatic Compounds. Huang, J.* and Mabury, S. A., University of Toronto, Toronto, ON M5S 3H6, Canada. Carbonate radical (CO_3^-) is a secondary radical produced from the scavenging of hydroxyl (OH) by carbonate/bicarbonate and is itself strongly electrophilic. As a selective oxidant, CO_3^- may be important for limiting the persistence of a number of aqueous pollutants, especially electron-rich aromatic sulfur compounds. Thioanisole, dibenzothiophene, and fenthion were selected as probes to determine the reaction pathway with CO_3^-. Using HPLC, GC, GC-MS and LC-MS for structural confirmation, the major photodegradation product of thioanisole, and dibenzothiophene was the corresponding sulfoxide, indicating the reactivity of CO_3^- towards sulfur. The sulfoxide products were further oxidized through reaction with CO_3^- to yield the corresponding sulfone products. Direct photolysis in DI water showed only slow conversion to the corresponding sulfoxide and sulfone products. Fenthion appeared to yield more degradation products due to both photolysis and hydrolysis; fenthion sulfoxide was the major product of reaction with CO_3^-. The identification of other metabolites and hydrolysis products of fenthion will be discussed. Results indicate CO_3^- may be an important contributor to the photo-induced degradation of aromatic sulfur-containg pesticides.

PWA179 Susceptibility of Intertidal and Subtidal Organisms to Photoenhanced PAH Toxicity. Pelletier, M.C.*, U.S. EPA, Narragansett, RI, Burgess, R.M., U.S. EPA, Narragansett, RI, Serbst, J.R., U.S. EPA, Narragansett, RI, May, A., Yale University, New Haven, CT, Cantwell, M., U.S. EPA, Narragansett, RI, Ho, K.T., Narragansett, RI. Phototoxicity is defined as a large increase in toxicity due to activation of polycyclic aromatic hydrocarbons (PAHs) by ultraviolet (UV) light. This phenomenon has been seen in both freshwater and marine organisms. In aquatic environments phototoxicity only occurs if UV light penetrates the water, as is found in shallow and intertidal areas. Deeper subtidal organisms are not exposed to sunlight under normal conditions. Intertidal organisms may have mechanisms to cope with UV exposure (i.e., pigmentation, shells, thicker cell walls) that would decrease their susceptibility to PAH phototoxicity. Subtidal organisms may not have these mechanisms and may be more susceptible. The main objective of this research is to determine the relative sensitivity of organisms to PAH phototoxicity based on ecological position (i.e., ultraviolet exposure, behavior, protective coverings). Several marine intertidal and subtidal species were collected from the field and exposed to fluoranthene for four days in a flow through system followed by one day of UV or fluorescent light exposure. Some organisms such as intertidal fiddler crabs, Uca pugnax, showed no PAH toxicity under UV light while the subtidal fish, Fundulus heteroclitus, exibited high toxicity. Bivalves living in the subtidal showed some PAH toxicity under UV light because their shells provided some UV protection. Correlation analysis of organism susceptibility with field measured ultraviolet intensities as well as habitat preferences and protective mechanisms suggest that toxicity by habitat type can be successfully modeled. This work will be useful in determining ecological risk in PAH contaminated areas.

PWA180 Phototoxicity of Anthracene and Fluoranthene in Killifish (*Fundulus heteroclitus*) from PAH-impacted and Non-impacted Sites. MacLean, E.D.*, Carey, J.A., Di Giulio, R.T., Duke University, Durham, NC. Polycyclic aromatic hydrocarbons (PAH) are carcinogenic and some are highly phototoxic to aquatic organisms. A population of killifish (*Fundulus heteroclitus*) has been identified in the highly PAH-contaminated Elizabeth River (ER), that, while exhibiting elevated rates of hepatic neoplasia, have been reported to be resistant to the acute and chronic toxicities of ER sediments relative to killifish from uncontaminated sites. Additionally, this resistance has been reported to be to be genetic. Given this background, we hypothesized that killifish from the ER are more resistant to the phototoxicities of anthracene and fluoranthene than killifish from uncontaminated sites. For these experiments we used age-matched laboratory-raised F1 offspring of killifish collected from the ER and two reference sites, York River (YR), VA, and Beaufort Inlet (BI), NC. Fish were exposed to anthracene and fluoranthene in the presence and absence of direct sunlight. LC_{50}s varied as a function of time for sunlight exposure and ranged from 37 ug/L at 40 minutes to 5ug/L at two hours for ER fish, and 60 to 15ug/L for the same time frame for BI fish. Similar differences were observed for anthracene exposure. Thus our results run directly counter to our hypothesis. This suggests that PAH toxicity may not be an important stressor for killifish inhabiting the ER, and that other mechanisms of toxicity dominate in this system, and while ER fish may have developed genetic resistance to some forms of toxicity, they may be more sensitive to others compared to fish from uncontaminated sites.

PWA181 An Assessment of Metal Contamination by Some Molluscs in Korean Coastal Waters. Koh, C.-H; Kang, S.-G.; Oh, I.S.; Lee, J.H.; Lee, C.H., Dept. of Oceanography, Seoul National University, Seoul 151-742, Korea. Cadmium, lead, copper and zinc concentrations in the soft body of the mussel (*Mytilus edulis galoprovinciallis*), oyster (*Crassostrea gigas*) and periwinkle (*Littorina brevicula*) were determined to assess the metal contamination in Korean coastal waters. Survey was carried out to describe the geographical trend in the metal content of these molluscs over the total of 50 stations along the Korean coast in January 1997. The metal concentrations varied highly with the sampling stations. The higher values of metal concentrations in the periwinkle were found in the Onsan Bay where an industrial complex is intensively developed. In other locations, metal values in the molluscs were at the natural background level. It indicates that the metal pollution in Korean coastal waters is not so critical except Onsan Bay. An intensive survey in Onsan Bay was, therefore, carried out to determine the degree of metal contamination and to find the input source of these metals within the Bay in November 1997. Surface seawater, mussels and periwinkles were taken at 20, 8 and 12 stations, respectively. The dissolved metal concentration in seawater was highest at the station near Onsan Nonferrous Industrial Complex, as being 1.15 μUg L^{-1} for Cd, 2.49 μUg L^{-1} for Pb, 3.75 μUg L^{-1} for Cu and 23.98 μUg L^{-1}. The metal values in the soft bodies of mussel and periwinkle from Onsan Bay were markedly higher than those from other locations of Korea. In the case of Cd, the highest values were recorded as 9.15 μUg g^{-1} by mussel, 27.1 μUg g^{-1} by periwinkle. Metal contents in the mussel and periwinkle showed a strong relationship with those in seawater.

PWA182 Biochemical and Physiological Responses of Saltmarsh Plants to Mercuric Chloride and Environmental Factors. Wall, V.D.*, Jacobs, H.E., Eastwood, A.L.; Moore, D.J., Pennings, S.C., University of Georgia Marine Institute, Sapelo Island, GA. Newly emergent *Spartina alterniflora* plants were acclimated to hydroponics in an open-sided greenhouse and exposed to $HgCl_2$ for 5 days. Photosynthesis (PS), transpiration (TS), peroxidase activity (POD) and total glutathione concentration (tGSH as GSH + GSSH) were measured. PS and TS levels were significantly lower than controls in the 10 ppm treatment group ($P<0.05$). POD and tGSH were significantly greater than controls in the 0.01, 0.1 and 10 ppm and 0.1, 1.0 and 10 ppm treatment groups ($P<0.05$), respectively. Results from a similar experiment with Juncus roemerianus were not dose-dependent but significant elevations of PS and TS were observed in the 0.1 ppm $HgCl_2$ treatment group. A two-way factorial (3x3 levels) experimental design with S. alterniflora was employed to determine the effect of $HgCl_2$ in the presence NaCl. Generally, increased salinity elevated levels of POD and tGSH and decreased PS and TS. $HgCl_2$ effects were less observable at higher salinities Sampling of *in situ S. alterniflora* at sites and times which bracketed natural environmental gradients indicated time of day and tidal inundation influenced tGSH and that unknown factors associated with spatial variation at high marsh elevations influenced POD. These results suggest that peroxidase activity and total glutathione concentration may be useful as biomarkers of $HgCl_2$ exposure in *Spartina alterniflora* as long as potential environmental influences are controlled for.

PWA183 Effect of Natural Organic Matter and Estuarine Salinities on Toxicity of Copper and Mercury. Pattanayek, M.*, Takács, M., Alberts, J.J., University of Georgia Marine Institute, Sapelo Island, GA. Laboratory tests were conducted to evaluate the effects of salinity and natural organic matter concentration on the potential toxicity of two heavy metals, i.e. Cu and Hg, as determined by Microtox® testing protocols. This study is part of a larger field experiment in southeastern U.S. salt marshes, which is investigating the potential use of salt marsh indigenous species as bioindicators of ecosystem condition. Conditions of the laboratory tests were designed to encompass the range of salinities (0-25 °/oo) and dissolved organic concentrations (10-30 ppm C) expected in these ecosystems. Natural organic matter was isolated from southeastern surface waters by ultrafiltration (nominal molecular weights >10,000 daltons) and characterized for C,H,N and ash content, fluorescence and FTIR spectral properties and total exchangeable proton and metal binding capacities. Chemical speciation of the test solutions were estimated using MINTEQ. Reduced toxicity (EC_{50}) for both copper (up to 92% decrease) and mercury (up to 46% decrease) was observed with increasing salinity and increasing organic matter content. The effect of photo-reaction on the organic matter and its effect of toxicity is also reported.

PWA184 The Effect of Mercury and PCBs on the Health of Organisms from Lower Trophic Levels of a Georgia Saltmarsh. Wall, V.D.*, Newell, S.Y., Pennings, S.C., Kneib, R.T. and Alberts, J.J., University of Georgia Marine Institute, Sapelo Island, GA. *Spartina alterniflora* dominated saltmarshes are among the most productive ecosystems in the world but little information is available on how pollutants affect these detrital-based food webs. Our approach to measuring the health of saltmarsh ecosystems includes rapid, inexpensive methods which assess the status of primary producers, fungal decomposers, bacterial populations and grass shrimp reproduction. Briefly, the methodology involved collection of sediment cores (for chemical analysis) and biological samples from a mercury/PCB contaminated site (LCP Superfund site) and a reference site (Turtle River). Primary production was assessed by measuring peroxidase activity, glutathione concentration, and rates of photosynthesis and transpiration. Microbial populations were assessed by measuring living-fungal standing crop (as ergosterol concentration), fungal sexual productivity, (as ascospore output) and Microtox®. Grass shrimp, *Palaemonetes pugio*, reproductive potential was determined by measuring brood size, brood mass, individual egg mass and average egg area. Comparison of the two sites using ANOVA procedures indicated that the mercury and PCBs present at the LCP Superfund site did not negatively affect the organisms we investigated. The results also suggested that due to natural environmental gradients, the chosen reference site wasn't an ideal control for the LCP site. Therefore, data from just the LCP site (excluding grass shrimp) were reanalyzed using multiple regression procedures. Significant relationships between methyl mercury in the sediment and both living-fungal standing crop and *S. alterniflora* peroxidase activity were observed, but natural environmental gradients confounded these relationships.

PWA185 Pilot Study for Synoptic Water Quality Monitoring in an Urban/Agricultural Watershed. Downing, J. W.*; Fairey, R.; Roberts, C.A. Moss Landing Marine Laboratories, Moss Landing, CA; Clark, R.P. California Coastal Commission, San Francisco, CA; Hunt, J.; Anderson, B.; Tjeerdema, R. University of California, Santa Cruz. Repeated high measures of pesticides and toxic response by amphipods in sediments from Moss Landing Harbor prompted study of pollutant sources in the watershed. Sediment and water were tested at seven stations in the lower reaches of the Tembladero watershed (Monterey County, California, USA) as a pilot study for monitoring many such urban/agricultural drainages in the region. Stations were selected at confluences to the Tembladero slough above where it empties into the Harbor at the Sandholdt Bridge. Chemical and toxicological tests were done, including sediment metal and organic chemistry analyses, water nitrate and TSS analyses. Water was tested for presence and relative concentrations of organochlorine pesticides using semipermiable membrane devices (SPMDs). Bulk phase sediment (*Eohaustorius estuarius* or *Hyalella azteca*) and water column (*Ceriodaphnia sp.* or *Holmesimysis costata*) toxicity tests were done at all stations. The most abundant metals were chromium and nickel, and the most abundant pesticides were Dieldrin, DDT, and Chlordane. The highest pesticide values (SPMD and Sediment) and strongest toxic responses were measured at the most upstream station. In general, water was toxic higher in the watershed, while sediment exhibited toxicity lower in the watershed. Results suggest that the watershed is acting as a sink as well as a source of pollutants for the Moss Landing Harbor. Recommendations for further investigation will be discussed.

PWA186 Risk Assessment for Dioxin Equivalents in Fish-eating Birds of Galveston Bay, TX. Cholger-Blust, S.L., Blankenship, A.L., Kannan, K., Giesy, J.P., Michigan State University, East Lansing, MI; Frank, D., Mora, M.A., U.S. Geological Survey, College Station, TX; Sericano, J., Wainwright, S., Texas A&M University, College Station, TX. Galveston Bay, TX, an inlet of the Gulf of Mexico, is a highly industrialized area and home to many species of fish-eating birds, including great egrets, black-crowned night herons, and cormorants. An ecological risk assessment of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents (TCDD-EQs) in birds of Galveston Bay was conducted to determine the likelihood of risk to native and migratory populations of fish-eating birds. TCDD-EQs were determined in egg extracts of these birds by an *in vitro* bioassay utilizing H4IIE-luciferase cells, which respond specifically to chemicals which bind to and activate the Ah receptor. TCDD-EQs ranged from below 11 pg/g (wet weight) to a maximum of 385 pg/g (wet weight). In addition, concentrations of individual polychlorinated dibenzo-*p*-dioxins (PCDDs), dibenzofurans (PCDFs) and polychlorinated biphenyls (PCBs) were determined by high resolution GC/MS. The relative contribution of each class of compounds to total TCDD equivalents (instrumental TEQs) was determined by multiplying congener-specific concentrations times toxic equivalency factors (TEFs) of the World Health Organization. A mass balance was performed to determine if all of the bioassay-derived TCDD-EQs could be accounted for by the TEQs measured by instrumental analysis. Maximum acceptable toxicant concentrations (MATCs) for TEQs in eggs, were derived by a combination of probabilistic and safety factor approaches. The probability of adverse effects of TEQs on the various species was determined by calculating hazard quotients (HQs) and then comparing the probabilities of exceedences for exposure and hazard.

PWA187 Composition and Source Identification of Hydrocarbons in Coastal and Offshore Sediments and Marine Animals In Areas of Oil/Gas Production Operations In the Gulf of Mexico. Sauer, T.C.*, Woods Hole Group, Falmouth, MA; Neff, J.M., Battelle, Duxbury, MA. Spatial and temporal trends in composition and sources of hydrocarbons in sediments and tissues of marine animals were evaluated near oil/gas production facilities in state and federal waters of the northwestern Gulf of Mexico. Produced water discharges were monitored at four offshore production facilities in 6 to 100 m of water and three nearshore facilities in Louisiana . The coastal sites were sampled before and after termination of produced water discharges. Saturated hydrocarbons (SHC: n-alkanes from C14 to C36 and isoprenoids), total hydrocarbons (THC), and polycyclic aromatic hydrocarbons (PAHs: over 40 alkylated PAH groups and priority pollutant PAHs) were determined in produced water, sediments, and marine animals from the immediate vicinity of each production facility. Sophisticated diagnostic parameters and multivariate statistical analyses, based on the SHC and PAH compositional fingerprints, were used to explain hydrocarbon composition signatures and differences in potential sources. Offshore, only the sediments nearest the four facilities contained petrogenic signatures which were superimposed on chronic industrial contamination signatures from the region, presumably from the Mississippi River discharge. Petrogenic PAHs were detected in edible tissues of bivalve mollusks and crabs from the vicinity of the platforms, especially near the shallow water offshore facilities. Shrimp and most fish samples did not contain petrogenic PAHs (<2 ng/g). Pyrogenic, not petrogenic PAH assemblages, were dominant in sediments near nearshore facilities. Sediments nearest these facilities, especially before discharge termination, had clear petrogenic PAH fingerprints. The use of only total PAH and THC data to assess oil/gas operational discharge impacts may lead to misinterpretations. Correct impact and recovery analysis requires use of source identification compositional methods. This work was sponsored by the U.S. Dept. of Energy.

PWA188 The Spatial Distribution of Polycyclic Aromatic Hydrocarbons in Sediments along an Estuarine Landscape Gradient in Coastal Regions of South Carolina. G. Scott, M. Sanders*, M. Fulton, S. Sivertsen NOAA/NOS Charleston Laboratory and J. Kucklick, NOAA/NIST, Charleston Laboratory, F. Holland, South Carolina Dept. of Natural Resources, and D. Sanger, Marine Science Program, University of South Carolina. Urbanization of the coastal zone of South Carolina has resulted in significant sources of polycyclic aromatic hydrocarbons (PAHs) including transportation corridors, marinas and residential areas. Nonpoint source runoff of PAHs from these sources may result in significant contamination of sediments in estuarine ecosystems. In addition, atmospheric deposition may be the primary source of PAHs in undeveloped and pristine habitats (e.g. NOAA National Estuarine Research Reserves and Sanctuaries Sites). The spatial distribution of PAHs in estuarine ecosystems has not been well documented in most national monitoring programs, particularly the difference in PAH levels in sediments from different estuarine microhabitats such as tidal creeks, tidal rivers, bays and sounds. A total of 210 station were sampled throughout an estuarine landscape gradient in Charleston Harbor which included small headwater tidal creeks, tidal rivers, sounds and bays. At each site sediments were collected from the top 10 cm of sediment and analyzed for 26 priority pollutant PAHs by High Performance Liquid Chromatography and Gas Chromatography-Mass Spectrometry. Results indicated that highest PAH sediment concentrations were found in small tidal creek habitats, particularly in highly urbanized watersheds. Sediment PAH concentrations were greatly reduced in rivers, bays and sounds. Comparison of sediment PAH concentrations with sediment quality guidelines (ERLs/ERMs) indicated that in tidal creeks, more than 50% of the stations had potentially toxic concentrations of PAHs. These high levels of PAHs may pose significant toxicological risks to larval/juvenile stages of fish and shellfish which use tidal creek habitat as nursery grounds during early potions of their life history development.

PWA189 **Vulnerability of the North Carolina Outer Coastal Plain to Contaminant Stress: Risk Assessment and Field Evaluation.** McCarthy, A.M.*, Cope, W.G., Shea, D., North Carolina State University, Raleigh, NC. The biological productivity of the Albemarle-Pamlico Sound, the second largest estuarine system in the United States, has declined substantially over the past 30 years. The National Oceanic and Atmospheric Administration published a hazard-based risk assessment for pesticides in 1992, which concluded that the outer coastal plain of North Carolina had the greatest risk of adverse ecological effects than any other region of the country, based on hazard normalized application rates. However this risk has not been empirically evaluated through field measurements. We assessed the ecological vulnerability of the North Carolina outer coastal plain to contaminants through field studies and chemical fate and effect models. The Pasquotank River Basin, which served as the model basin in our study, has two fish consumption advisories (one for dioxin and the other for mercury), has relatively high pesticide application rates, and is undergoing rapid development. Mercury concentrations in surficial sediments, fish and shellfish ranged from 0.15 to 47 ppm. Polychlorinated biphenyls concentrations in fish range from 0.06 to 5.2 ppm wet weight. The Pasquotank Basin was used as a model system to determine the hazards of exposure to chemical mixtures and metals in the watershed. Sediment, water and biota were collected from sites in the basin and have been used to evaluate the risk of both currently used pesticides and historically persistent contaminants. Data gathered has been used to develop a fate model for the region in an attempt to determine if the decline in biological productivity is linked to contamination.

PWA190 **Anthropogenic Nitrogen Pollution in the Neuse River-Estuary, NC: Effects of Nitrogen Compound Compositions on Phytoplankton Community Dynamics.** Harrington, M. B.*, Pinckney, J. L., Paerl, H. W.; UNC Institute of Marine Sciences, Morehead City, NC. The Neuse River-Estuary, NC is a chronically nitrogen-limited system experiencing eutrophication due to increasing nitrogen (N) loading to the estuary. "N is N" is a common assumption when relating nutrient inputs to structural and functional alterations of natural phytoplankton communities. However, N inputs can come from many sources which supply chemically distinct N compounds, including nitrate, ammonium, and organic N. These compounds have different bioreactivities which may lead to physiologically distinct responses in phytoplankton communities. The relative proportion of ammonium in riverine and atmospheric N inputs to the Neuse River watershed and estuary is currently increasing. To determine if this change in N supply causes a corresponding compositional (and accompanying functional) shift in the local phytoplankton community, we performed small-scale (4 liter) manipulative bioassays. Community responses (primary productivity, biomass, algal composition) to nitrate and ammonium additions were measured over 48 hours. Preliminary statistical analysis of results (using Bonferroni tests) showed that the phytoplankton community was N-limited. Primary productivity ($p<0.001$) and total biomass ($p<0.01$), measured as concentrations of chlorophyll *a*, both responded significantly to N additions. Certain algal groups (as evidenced by biomarker pigment concentrations) also responded to N additions, including cryptomonads ($p<0.005$ in 100% of cases) and diatoms ($p<0.02$ in 83% of cases). There were no significant differences in response of biomass, productivity, or algal group composition to different N compounds.

PWA191 **Will nutrient management strategies control phytoplankton overproduction in the Neuse River-Estuary NC, USA?** Piehler, M.F.*, Pinckney, J.L., and Paerl, H.W.; UNC-CH Institute of Marine Sciences, Morehead City, NC. The Neuse River-Estuary has experienced rapid and significant deterioration in water quality in recent years. Reduction in nitrogen (N)-loading is recognized as an effective means to control nuisance algal growth, but the magnitude of decrease in production by N reduction has not recently been thoroughly assessed. Dilution bioassays were employed to predict effects of proposed 30% N-loading reduction on native Neuse River-Estuary phytoplankton productivity ($^{14}CO_2$ incorporation) and biomass (Chl *a*). Samples were taken from a riverine and an estuarine site. Treatments included simulation of reductions in concentrations of N, phosphorus (P), and N&P. Bioassays were run throughout the year to account for seasonal variance (August, October, January, March). Mean Chl *a* concentration remained below the level of the control throughout the five day incubations for both sites in all but one experimental simulation of a 30% N reduction (August for the riverine, January for the estuarine). Following 30% N dilution, assimilation number (productivity/Chl *a*) was reduced below the level of the control in two of four times at the estuarine site and three of four at the riverine site. Generally, dilution of N&P reduced productivity and standing stock the largest amount, followed by N reduction alone and then P reduction alone. These results indicate that reducing N loading to the Neuse River-Estuary by 30% will likely decrease both phytoplankton biomass and primary productivity throughout the majority of the year. Reducing both N and P consistently led to the largest decreases in biomass and productivity, suggesting a synergy of parallel N and P reductions.

PWA192 **The Flux and Bioavailability of Atmospheric Organic Nitrogen to North Carolina Coastal Marine Ecosystems.** Peierls, B.P.* and Paerl, H.W., UNC-CH Institute of Marine Sciences, Morehead City, NC. Atmospheric nitrogen (N) deposition contributes to the overenrichment of N-limited marine waters which can lead to nuisance or toxic algal blooms, hypoxic or anoxic conditions, community composition shifts, and other symptoms of eutrophication. Atmospheric dissolved organic nitrogen (ADON) has recently gained attention as a significant additional, yet rarely assessed, source of atmospheric N loading to coastal and estuarine ecosystems. Little is known, however, about the potential for ADON as a nutrient source for primary producers. The flux and potential bioavailability of ADON in coastal North Carolina was evaluated using event-based collections and enrichment bioassays. A high-temperature oxidation technique applied to two years of rainwater samples revealed an average DON concentration of from 2 to 4 μM, or approximately 10-20% of total N concentration. Annual wet DON deposition was 11 % of total N deposition and was greatest for season and storm types with the most rainfall. Enrichment of coastal water with isolated rainwater DON produced increased phytoplankton biomass and productivity that ranged from 20-60% of the response to inorganic N additions. The bioassay results suggest that as much as one-half of the ADON pool is available to primary producers on short (hours to days) time scales. The impact of ADON on marine ecosystems over longer time scales and at natural loading rates requires further investigation.

PWA193 **The status and trends of trace element and organic contaminants in oysters, *Crassostrea virginica*, in the waters of the Carolinas.** Lauenstein, G. G. and T. P. O'Connor. National Oceanic and Atmospheric Administration, National Status and Trends, Silver Spring, MD. Concentrations of ten trace elements (Ag, As, Cd, Cr, Cu, Hg, Ni, Pb, Se, Zn), lindane and six groups of organic contaminants (chlordane, total-PCB, total-DDT, Dieldrin, butyltins, PAHs) at the 11 NOAA Mussel Watch Project (MWP) sites located in North and South Carolina have been compared with the national MWP data. Three sites from North and South Carolina had concentrations of PAHs in the upper 15th percentile on a national scale, one site had high concentrations of butyltins, and two sites had high Se concentrations. All sites from Beaufort, North Carolina south had high As concentrations. Temporal trends were calculated for all chemicals at all sites. Decreasing trends were found for Ag, As, Cd, chlordane, total-DDT, total-PCB, and PAHs at some sites, though no one site had decreasing trends for all analytes.

PWA194 Polychlorinated Naphthalenes (PCNs) in Soil, Sediment and Biota Collected Near a Former Chlor Alkali Plant in Coastal Georgia. Kannan, K.*, Blankenship, A., Giesy, J.P., Michigan State University, East Lansing, MI; Imagawa, T., National Institute for Resources and Environment, Tsukuba, Japan. Isomer-specific concentrations of polychlorinated naphthalenes (PCNs) were determined in soil, sediment, striped mullet and boat-tailed grackle collected near a chlor alkali plant in coastal Georgia to examine congener profile, bioaccumulation and toxic potential. PCN concentrations as great as 23 µg/g, dry wt, were found in sediments collected at the marsh contaminated by the disposal of wastes from chlor alkali process. Spatial distribution of PCN concentrations in the marsh was not correlated with those observed for polychlorinated biphenyls. The PCN congener profile did not resemble to those in any of the technical mixtures. Hepta- and octachloronaphthalenes were the most abundant congeners accounting for >50% of the total PCN concentrations in soil and sediments. The concentrations of PCNs in fish and birds were 3- to 5-orders of magnitude less than in sediments, and dominated by tetra- or pentachloronaphthalenes. Affinity of planar, more chlorinated naphthalene congeners to sediment organic carbon and steric factors that affect membrane permeability have contributed to less bioavailability and bioaccumulation. The 2,3,7,8-TCDD equivalents (TEQs) estimated for PCNs in sediments and biota were greater than those reported for PCBs, polychlorinated dibenzo-p-dioxins (PCDDs) or dibenzofurans (PCDFs). Our results suggest that chlor alkali process has been an important source of PCNs, both due to their formation during the process and the use of technical PCN mixtures as lubricants in the electrodes. Contribution of PCNs to dioxin-like toxicity in environmental media nearby chlor alkali process may overwhelm those due to PCBs, PCDDs or PCDFs.

PWA195 PAH Effects on Benthic Community Structure. Martin, M.M.*; Oberdorster, E., Tulane/Xavier Center for Bioenvironmental Research, New Orleans, LA. The impacts of polycyclic aromatic hydrocarbon (PAH) and lead contamination on benthic community structure were investigated in Bayou Trepagnier (BTP) in Southeastern Louisiana. BTP has been heavily contaminated since the 1920s by the Petroleum Industry with tar sludge and lead. Previous studies have shown that PAH and heavy metal contamination within Bayou Trepagnier is extremely patchy. The objectives of this study were to link benthic community structure to PAH and heavy metal contaminant levels, and assess diversity between more and less contaminated site within Bayou Trepagnier. Preliminary studies showed that variability due to ecological differences between potential control bayous and BTP would mask any contaminant effects. Comparing cleaner and more contaminated sites within Trepagnier, differences due to ecology were minimized. Macrofauna and meiofauna were sampled from BTP sites 61,71,96 and 111. Cores were taken in triplicate to two cm depth with 5.3cm2 syringe corer, and samples were fixed in ten percent formalin with Rose Bengal, seived, and separated for macrofauna (0.5mm mesh size) and meiofauna (0.063mm mesh size). The stained sediment was counted using a Buggeraff tray, and number of individual per Phylum or Class and number of identifiable species were noted for each sample. Data was calculated as animals/m 2 and number of species for each site. The results of the study showed an overall low diversity and low abundance of macrofauna, which is common for bayous in Southeastern Louisiana. Sites with highest contaminant levels (BTP111 and 71) had significantly lower number of nematodes, oligochaetes, rotifers and total number of animals for both meiofauna and macrofauna. Diversity was significantly decreased only for meiofauna at site 111. In Bayou Trepagnier, there is a significant decrease in macrofaunal and meiofaunal benthic communities along a PAH and lead contamination gradient.

PWA196 Biological and Ecotoxicological Characteristics of Terrestrial Vertebrate Species Residing in Estuaries. Golden, N.H.*, Pearson, J.L., Ottinger, M.A., University of Maryland, College Park, MD; Rattner, B.A., Erwin, R.M., USGS Patuxent Wildlife Research Center, Laurel, MD. The threat of contaminants to terrestrial vertebrates residing in Atlantic coast estuaries is being examined by the Department of the Interior's Biomonitoring of Environmental Status and Trends program. Twenty-two species (2 reptiles, 18 birds, 2 mammals) that breed in proximity to estuaries were evaluated for potential suitability as sentinels of environmental contamination. Species were selected based upon documented or suspected contaminant exposure and effects, or because they are valued or protected natural resources. The morphology, status in estuaries, abundance and range, site fidelity, ease of census, feeding habits, and a summary of published contaminant exposure and response data for each species was compiled by review of extant data. Data are being nonparametrically analyzed to rank the suitability and sensitivity of the select species for monitoring various classes of contaminants and estuarine health. Preliminary findings indicate substantial chlorinated hydrocarbon exposure data for 10 of the 22 candidates, most of which are piscivorus and colonial breeding waterbirds that readily bioaccumulate these compounds. Based on reproductive effects (population declines and recoveries), the bald eagle, brown pelican and osprey appear to be the most sensitive to chlorinated hydrocarbons. There are considerably less metal exposure data, and almost no associated effects data. The most substantial metal data (lead shot and sinker ingestion) are for mute swan and black duck, species that feed by grazing or dabbling. Despite numerous incidences of organophosphorus and carbamate poisoning in birds, published reports fail to reveal any single estuarine vertebrate species or group as an outstanding sentinel of anticholinesterase intoxication. Limited field data are available for petroleum hydrocarbons, although vulnerability studies suggest diving ducks are most susceptible.

PWA197 Ecological Risk Assessment for Restoration of the Bolsa Chica Lowlands, California. Ohlendorf, H.M.*, Castleberry, M.A., CH2M HILL, Sacramento, CA; Kinney, P.K., Stevenson, M.L., Kronschnabl, K.P., Kinnetic Laboratory, Santa Cruz, CA; Rivera, M., U.S. Fish and Wildlife Service, Carlsbad, CA. The main focus of this complex, large project is to conduct sampling and to perform an ecological risk assessment for the Bolsa Chica Lowlands in Orange County, CA, which includes about 485 hectares (1,200 acres). This work is being conducted to characterize contamination within the Lowlands and to establish cleanup criteria for portions of the property affected by previous activities, primarily oil and gas production and urban runoff. More than 430 oil wells exist on the property (including many abandoned wells), along with associated pipelines, roads, former tank farms, and other related facilities. Storm drainage enters from nearby urbanized areas. Once the full extent of contamination is understood and appropriate risk assessment-based cleanup levels have been established, it is expected that contaminated areas will be remediated and the Lowlands will be restored to provide a mix of tidal coastal wetland habitats and non-tidal seasonal ponds. The overall project includes a wide range of federal and state agencies, as well as private organizations, having an interest in restoration of the Lowlands as mitigation for other construction projects, with the U.S. Fish and Wildlife Service serving as the contracting agency.

PWA198 Non-target Effects of the Mosquito Larvicides, Temephos and Methoprene, at Bombay Hook and Prime Hook National Wildlife Refuges. Pinkney, A.E.*, McGowan, P.C., Murphy, D.R., U.S. Fish and Wildlife Service, Annapolis, MD; Sparling, D.W., Lowe, T.P. Patuxent Wildlife Research Center, Laurel, MD; Ferrington, L.C., University of Kansas, Lawrence, KS. The U.S. Fish and Wildlife Service is currently evaluating the environmental risks associated with mosquito control procedures at National Wildlife Refuges to ensure their compatibility with refuge objectives and operations. As part of that effort, we investigated the non-target effects of temephos (applied as Abate 4E) and methoprene (applied as Altosid Liquid Larvicide) on the salt marsh and on freshwater habitats. Three test plots at Bombay Hook NWR (Delaware) were sprayed on four occasions with Abate 4E at 0.054 kg active ingredient/hectare while three were unsprayed. There were no statistically significant effects on populations of marsh fiddler crabs (*Uca pugnax*) based on counting of burrow holes and *in situ* bioassays. Spraying the test plots with Altosid at 0.011 kg a.i./ha did not result in a statistically significant effect on fiddler crab reproduction as measured by the percentage of females that were gravid. Three groups of six manmade ponds at Patuxent Wildlife Research Center were sprayed with either Abate, Altosid, or distilled water on three occasions at three week intervals. The emergence of insects was monitored before and after each spray. There were significant decreases in the mean number of individuals, species, and families in the Abate ponds relative to the controls. Species diversity and equitability and the mean numbers of Ephemeroptera, Odonata, Chironomidae, and Chaoborus were significantly lower in the Abate ponds relative to the control ponds. There were no effects in the ponds sprayed with Altosid. Management recommendations include continued use of the larvicides on salt marsh habitats with restricted use on freshwater habitats.

PWA199 Duwamish Estuary Ecological Risk Assessment: Modeling Exposures from CSOs and Other Sources. K. Schock, B. Swarner and J. Zhong, King County Department of Natural Resources, J. Wilbur, The Boeing Company, Inc., J. Hamrick, Tetra Tech, Inc., R. Walton, WEST Consultants, Inc., and J.Toll*, Parametrix, Inc. The Duwamish River and Elliott Bay are important natural and economic resources for the Puget Sound region. Located in Seattle, Washington, they form a heavily used urban estuary and an important ecological environment. The estuary is well stratified (salt-wedge type) and the position of the wedge toe is very dynamic, being controlled by tides and fresh-water flow. King County Department of Natural Resources selected the three-dimensional Environmental Fluid Dynamics Computer Code (EFDC) to model density flow and toxicant transport and fate within the estuary and bay. The study area was segmented into 500 cells in the horizontal plane, and ten layers in the vertical for 5,000 cells in total. The model was run on a Cray supercomputer provided by Boeing. EFDC was modified to simulate near-field CSO effects within the larger model cells. An intensive field sampling program was initiated to collect water and sediment chemical data to calibrate the toxicant transport and fate module, and physical data to calibrate the hydrodynamic module. Post-processing of the EFDC output was performed to extract the data required to assess risks to aquatic life, wildlife and people. This is an important modeling effort because it provides a sophisticated, generalized tool for making socioeconomic decisions about managing resources in the Duwamish River watershed.

PWA200 A Case Study: Data Compilation, Evaluation, and COI Selection for a Sediment-Focused Ecological Risk Assessment. K.K. Shearer*, T.R. Barber, E.A. Fendick, P.C. Fuchsman, S.A. Keenan, B.S. Peck; ChemRisk, Cleveland, OH. An Ecological Risk Assessment (ERA) focusing on potentially impacted sediments is in progress for a large industrial facility as part of a Resource Conservation and Recovery Act (RCRA) Facility Investigation (RFI). A total of 196 sediment samples, collected from ecologically relevant locations during 12 sampling programs spanning 6 years, were analyzed for target chemicals (SVOCS, VOCs, PAHs, pesticides, PCBs, and inorganics). Laboratory results (totaling over 22,000 records) were compiled in a relational database and prepared for further evaluation. Summary statistics were calculated to characterize concentrations of individual chemicals. Chemicals of Interest (COIs) are identified in sediment using selection procedures designed to focus subsequent ERA efforts on chemicals most likely to contribute to ecological risk, and to eliminate from quantitative analysis chemicals unlikely to contribute to risk. COI selection procedures consist of the following 6 screening steps: (1) a comparison of inorganic concentrations in site-related sediment to background concentrations derived from a regional sediment data set, (2) an evaluation of the potential for chemical concentrations to cause adverse effects (based on comparison to screening values from Federal sources or calculated from water quality benchmarks using equilibrium partitioning methods (EqP)), 3) status as an essential nutrient, (4) capacity to cause toxic effects (5) use in common laboratory procedures, and (6) determination of the frequency and location of chemical detections. Of the 88 chemicals detected in the 196 sediment samples, 30 are identified as COIs for further evaluation in the ERA. The poster outlines the processes involved in data compilation, evaluation, and COI selection for an ERA of a large industrial facility.

PWA201 Development of marine sediment toxicity data for ordnance compounds and toxicity identification evaluation (TIE) studies at select naval facilities. Carr, R.*, Biedenbach, J., and Hooten, R., USGS, Marine Ecotoxicology Research Station (MERS),Corpus Christi, TX; Nipper, M., TAMU-CC, Center for Coastal Studies, Corpus Christi, TX; Saepoff, S. Engineering Field Activity, Northwest (EFA NW), Poulsbo, WA; and Miller, K., Naval Facilities Engineering Service Center (NFESC), Port Hueneme, CA. Several sites in the vicinity of Naval facilities are suspected of being contaminated with ordnance compounds (i.e., 1,3,5-trinitrobenzene, 1,3-dinitrobenzene, picric acid, tetryl, otto fuel, 2,4,6-trinitrotoluene, 2,4,-dinitrotoluene, 2,6,-dinitrotoluene and royal demolition explosive) from past use, storage, improper disposal, and incineration of these compounds. Based on a comprehensive literature search, conducted by Oak Ridge National Laboratories (1996), on the available data on ordnance toxicity, little or no toxicity data for benthic marine or estuarine organisms and no Sediment Quality Standards (SQS) currently exist for these substances or their degradation products. Initially, sediment quality assessment surveys were conducted at several of the sites suspected of ordnance contamination. Based on the results of these initial toxicity and chemical surveys, several stations at each site were selected for sediment toxicity identification evaluations (TIEs) to ascertain whether ordnance compounds were responsible for any observed toxicity. In addition, a toxicity data base for these compounds was developed using a variety of sensitive marine organisms (e.g., sea urchin, *Arbacia punctulata*, fertilization and embryological development; polychaete, *Dinophilus gyrociliatus*, life-cycle test; fish, *Sciaenops ocellatus* and crab, *Callinectes sapidus*, embryolarval development; *Ulva* sp. zoospore germination and growth test). The information generated from these studies will be used to support the development of water quality criteria and sediment quality guidelines for use in a regulatory framework on a national level.

PWA202 Ecotoxicological and chemical characterization of sediment and dredged material from Santos Estuary, São Paulo/Brazil. Prósperi, V. A.*, Valent,G.U; Eysink, G.G.J.; Pompéia, S.L. CETESB-Environmental Sanitation Technology Agency, São Paulo, SP, Brazil. In harbor areas dredging is usually necessary and involves several environmental and economical problems. This activity may result in a variety of negative effects on the ecosystem, especially when the material is contaminated with anthropogenic pollutants. Santos harbor, the most important one of São Paulo State/Brazil, is located in an estuary, near Cubatão City, with a huge industrial complex, including steel, fertilizers, chemical and petroleum plants. The effluents have been discharged directly in this estuary for several years and as consequence the mangrove were degraded and the water, sediment and biota contaminated. In 1984, efforts were made in order to control the main pollution sources and it was observed an improvement in the water quality. Because of the intense harbor activities, more than 5,000 ton of sediment must be dredged every year, and until now, the material has been disposed in the ocean. The aim of this study was to perform a chemical and ecotoxicological characterization and the results showed that the sediment and dredged material itself were mainly contaminated with PAH's especially benzo(a)pyrene (up to $194x10^3$µg/Kg) and metals such as Hg (0,83µg/g), Ni (57µg/g), Cu (81µg/g), Pb (567µg/g) and Zn (2600µg/g). Some samples were also mutagenic for *Salmonella typhimurium* and impaired embryo development of *Lytechinus variegatus*. The Environmental Regulatory Agency (CETESB) interrupted the dredging activities of the most contaminated areas and at the same time started a detailed study in order to define criteria to properly classify, manage and dispose of the sediment.

PWA203 Assessment of Sediment Quality Effects in Mission Bay and San Diego Bay on Juvenile White Seabass (*Atractoscion nobilis*) and California Halibut (*Paralichthys californicus*). Stransky, B.C.*, California State University, San Diego, CA., Ogden Environmental and Energy Services Inc. and Ford, R.F. California State University, San Diego, CA. The objective of this study was to assess whether sediment contaminants in Mission Bay and San Diego Bay pose a substantial risk to juvenile fish species, specifically white seabass and California halibut, which utilize these areas as a nursery ground. Sediment collected from five sites in Mission Bay induced few adverse effects in either fish species during a 14-day exposure in flow-through aquaria. However, fin erosion did develop on a number of California halibut and white seabass after exposure to sediment from a back-bay Mission Bay location, Tecolote Creek Inlet. High levels of hydrocarbons were present at this site. Both fish species were also exposed to sediments from one industrialized region in San Diego Bay, Commercial Basin. High levels of hydrocarbons and the trace metals copper and mercury have been measured at this site. Relative to responses obtained after exposure to sediment from Mission Bay and the coastal surf zone, both species exhibited decreased growth rates and white seabass displayed weaker swimming performance and feeding responses. Juvenile California halibut also developed external lesions and displayed sediment avoidance behavior. The incidence of fin erosion among both species was similar to that found after exposure to sediment from Tecolote Creek Inlet. Results and observations during this study suggest that the substrate-oriented behavior of juvenile white seabass and California halibut render both of these species highly susceptible to contaminated sediments in San Diego Bay. The development of fin erosion among fish exposed to sediments from east Mission Bay may be of some concern and warrants further attention.

PWA204 Development of biomarkers using an *in situ* animal model to assess estrogenic activity in municipal wastewater. Tilton F.*, Schlenk, D., and Benson W.H. The University of Mississippi, University, MS. The estrogenic activity associated with effluent from two municipal wastewater treatment plants (WWTP) was assessed in the fall 1996-97 and the spring 1997. Fish were maintained at two sites within each WWTP; sites were located at pre-chlorination (P) and the post-chlorination stream (S). One WWTP had a retention time of 1 day (R1), while the second WWTP had a retention time of 60 days (R60). The sites were designated as: R1-pre-chlorinated (R1-P), R1-post-chlorinated stream (R1-S) and R60-P and R60-S, respectively. Mature male channel catfish (n=10; 200 to 300 g) were placed in cages at each experimental and reference (REF) site and exposed *in situ* for 21 days. Hepatic/somatic index (HSI) at the fall 1997 R60 sites were elevated 1.3-1.7 fold greater than REF values. Gonad/somatic indices (GSI) showed no significant differences in any sampling season. Levels of serum vitellogenin (Vtg), at the fall 1996 R60 sites, were 2.5 fold over REF values. In the fall 1997 R1-P site and all spring 1997 sites, serum Vtg levels where 4.8 fold and 3.4-3.8 fold greater than REF, respectively. Hepatic CYP1A content was significantly increased 1.4-10.1 fold greater than REF during both seasons except the fall 1997 R60-P site. Hepatic ethoxyresorufin O-deethylase (EROD) activity was significantly elevated greater than REF in a range of 2.6-4.1 fold at all the pre-chlorinated sites, except the fall 1997 R1-P site. EROD values for spring 1997 R60-S site were elevated 2.2 fold above REF.

PWA205 Impact of natural and synthetic estrogens in municipal effluents in Ontario. Servos, M.*, Bennie, D., Brown, S., Burnison, K., J., Sherry, A. Gamble, M. Hewitt, J., McInnis, R., Toito, J., Jurkovic, A., National Water Research Institute, Burlington, Ontario, Ternes, T., EWSE, Germany Schnell, A., Wastewater Technologies International, Burlington, Ontario, Van Der Kraak, G., University of Guelph, Guelph, Ontario, Canada. Natural and synthetic estrogens have been identified as trace contaminants in municipal effluents. These chemicals are suspected of causing a variety of biological responses in fish downstream of municipal treatment plant outfalls, including the induction of vitellogenin synthesis and intersex in fish. The prevalence and effects of these chemicals in municipal treatment systems in Canada has not been documented. Effluent samples from municipal treatment systems in Ontario were examined for natural and synthetic estrogens to determine their prevalence and potential release into the environment. At selected sites the concentrations of alkylphenol polyethoxylates, a group of known estrogenic contaminants in municipal treatment systems were also determined. Selected samples were also tested using the YES assay system. 17β-estradiol, estrone and ethenylestradiol were detected in most of the final effluents tested in the low ng/L range. Immature rainbow trout exposed to municipal effluents in the laboratory (21 d, 2 d renewal) from the same sites showed weak induction of vitellogenin in plasma. Rainbow trout caged downstream of the outfalls of several municipal treatment plants also showed weak vitellogenin induction after a three weak exposure. Additional samples of wild fish were collected and the gonads sectioned for histological examination. A national survey of municipal treatment plants is currently planned to assess the prevalence of these compounds entering aquatic environments in Canada.

PWA206 Runoff of estrogens into small streams after the application of hog manure to agricultural fields in southern Ontario. Servos, M., Brown, S., Burnison, K., Mayer,T., Parrott, J., Sherry, J., McInnis, R., Toito, J., Jurkovic, A. National Water Research Institute, Burlington, Ontario, Canada, E.*,Topp, Agriculture Canada, London, Ontario, Canada. Natural estrogens have been identified as a component of hog manures that are widely applied to agricultural fields in Ontario. These compounds can runoff of the fields and enter aquatic environments where they have the potential to cause a variety of effects on the endocrine function of fish and other biota. The extent to which these compounds enter aquatic environments and impact fish has not been well documented. To address this question samples of manure, soil, tile drainage and stream water were taken after the application of approximately 6,000 gal/acre of manure to three agricultural fields in south-western Ontario. The three hog production facilities studied included small (65 head) and large (1000 head) feed operations, and a sow (700 head) operation where the manure was stored over the winter in large lagoons and applied to the fields in the last week of April.. Samples were collected immediately after the applications as well as after a major rainfall event. Samples were examined for 17β-estradiol and estrone using GC/MSD and EIA methods and extracts were assayed using the YES system. Immature rainbow trout were caged for three weeks in the streams immediately above and below the fields and plasma was assayed for vitellogenin. Semi-permeable membrane devices (SPMD) were placed adjacent to the fish cages during the fish exposure period. Chemical and biological responses are compared at the three sites.

PWA207 Effects of Model Sewage Treatment Plant Effluents on Circulating Sex Steroids in Rainbow Trout. Parrott, J.L., McMaster, M.E., Jardine, J.J., National Water Research Institute, Environment Canada, Burlington, Ontario, Canada; Irwin, D., Steller, S., Livingstone, S., Trowbridge, D., Verma, S., Fruchter, J., Sault College, Sault Ste. Marie, Ontario, Canada; Shaw, M., Upper Lakes Environmental Research Network, Sault College, Sault Ste. Marie, Ontario, Canada. Results of fish exposed to sewage treatment plant (STP) effluents in the UK showed elevated vitellogenin, and evidence of intersex fish. To investigate whether Canadian sewage effluents had steroid-disrupting effects, a short-term test with rainbow trout was developed. Reproductive steroids were measured in immature (40-60 g) rainbow trout after a 21 day exposure to effluent from a model sewage treatment facility at Sault College, Sault Saint Marie, Ontario. Fish were exposed in garbage pails, with solutions renewed three times a week. Temperature was maintained by water baths at 9-11 °C. Fish survived exposures to 10 % primary treated sewage effluent, 100 % final sewage effluent and to high (348 ng/L) and low (174 ng/L) estradiol. Rainbow trout exposed to primary or final effluent showed five-fold elevations in blood testosterone. Exposure to estradiol had no effect on testosterone levels, compared to water or solvent (ethanol) control fish. Blood estradiol levels showed similar trends in primary and final effluent-exposed fish, but effects were slight (1.2 to 1.4-fold increase) and not significant. Estradiol treatment raised estradiol in blood of exposed trout, as expected. The result show that this model STP effluent can influence steroid levels in fish. As well, the rainbow trout static-renewal 21 day exposures proved to be a simple bioassay, easy to carry out on site, and the test appears promising for use in field locations.

PWA208 Effects of Endocrine-Disrupting Chemicals on Plasma Testosterone, 17b-Estradiol, Vitellogenin and Gonadosomatic Index in Juvenile Male Summer Flounder (*Paralichthys dentatus*). Gutjahr-Gobell, R.*, Borsay, D., Zaroogian, G., Haebler, R., Jayaraman, S., McKinney, R., Pruell, R., Mills, L. U.S. EPA, Narragansett, RI. Effects of endocrine-disrupting chemicals on the sexual development of juvenile male summer flounder, *Paralichthys dentatus*, were studied in the laboratory. Estrogenic (*o,p'*-DDT, octylphenol) and antiandrogenic (*p,p'*-DDE) chemicals were studied alone and in combination (*o,p'*-DDT+ octylphenol, *p,p'*-DDE+octylphenol). Test chemicals were dissolved in acetone, added to coconut oil, and then injected into the dorsal sinus (initially and again at two weeks) where it solidified into a bolus. A carrier-control (coconut oil) and positive-control (17b-estradiol) were included with each experiment. Blood and tissues were sampled at 4, 6 and 8 weeks. Fish exposed to *o,p'*-DDT+octylphenol were also sampled at 15 weeks. Compared to the carrier-control: 1) 17b-estradiol reduced plasma testosterone, elevated plasma estradiol, caused significant vitellogenin production and reduced gonadosomatic index (GSI) at 4, 6 and 8 weeks; 2) *o,p'*-DDT significantly reduced plasma testosterone at 4 weeks, didn't affect plasma estradiol, marginally induced vitellogenin and significantly reduced GSI at 4, 6 and 8 weeks. 3) octylphenol reduced plasma testosterone and elevated plasma estradiol at 4 weeks, didn't cause vitellogenin production, and reduced GSI at 4,6 and 8 weeks; 4) *o,p'*-DDT+octylphenol reduced plasma testosterone, increased plasma estradiol at 15 weeks, didn't induce vitellogenin and reduced GSI at 8 and 15 weeks; 5) *p,p'*-DDE+octylphenol reduced plasma testosterone, didn't affect plasma estradiol or induce vitellogenin and reduced GSI at

8 weeks; 6) *p,p* '-DDE didn't affect plasma testosterone, plasma estradiol, vitellogenin production or GSI. Flounder that were exposed to estrogenic chemicals exhibited effects on testosterone & 17b-estradiol production, vitellogenin induction and GSI while fish exposed to the antiandrogen did not.

PWA209 The Use of Steroid Hormones in Examining Endocrine Disruption: An Interlaboratory Study. McMaster, M.E.*, Jardine, J.J. and Munkittrick, K.R. Environment Canada, Burlington, Ontario, Canada. We have initiated an interlaboratory study to examine variability in the measurement of steroid hormone levels. In recent years, there has been an increased use of the measurement of steroid hormone levels in the blood of animals exposed to contaminants as an indicator of reproductive impairment. The most commonly used technique at present is the radioimmunoassay however, there are also a number of alternative analytical techniques. Although we are confident that significant reductions in steroid hormone levels can be detected when they exist using these techniques, there has been no study to examine the variability in these techniques or between laboratories. For this reason, during a recent field study examining fish downstream of a pulp mill for evidence of alterations in steroid hormone levels, we collected sufficient samples to conduct such an interlaboratory study. White sucker were collected from the exposed as well as a reference location and samples were taken for blood as well as *in vitro* steroid production by gonadal tissue. Samples were delivered to 11 different laboratories with the ability to measure steroid hormones. Samples were submitted blind and the ability of the labs to detect site differences was determined. Although the magnitude of steroid hormone content in the samples varied somewhat between labs, the majority of the laboratories were capable of identifying site differences in steroid levels. Labs with difficulties in steroid measurements were investigated in more detail to examine the problems.

PWA210 The Use of Testosterone Hydroxylase Activities as a Biomarker of Endocrine Disruption. Wilson, V. S.*, McLachlan, J.B., Falls, J. G., LeBlanc, G. A., North Carolina State University, Raleigh, NC. This study evaluated whether changes in androgen-regulated testosterone biotransformation processes could be used as a biomarker of exposure to androgen-disrupting chemicals. Sex specific differences in hepatic testosterone biotransformation enzymes were identified. Gonadectomy and hormone replacement experiments established that many of these sex differences were due to the inductive or suppressive effects of testosterone. Most notably, significant androgen-dependent differences in the ratio of testosterone 6a- and 15a-hydroxylase activities in both sexes were demonstrated with females having a significantly higher ratio than males. Next, experiments were conducted to determine whether this ratio could be used as a biomarker of exposure to androgen-disruption chemicals. Exposure of male mice to the antiandrogen, vinclozolin, significantly increased the 6a/15a-hydroxylase ratio. Male mice were also exposed to indole-3-carbinol and 4-nonylphenol that, respectively, reduced or increased serum testosterone levels. Indole-3-carbinol increased the 6a/15a-hydroxylase ratio, while 4-nonylphenol decreased the ratio. The 6a/15a-hydroxylase ratio proved to be a powerful measure of androgen modulation and a sensitive measure of exposure to androgen-disrupting chemicals in CD-1 mice.

PWA211 Plasma Steroid Levels in Kootenai River White Sturgeon (*Acipenser transmontanus*) as an Indicator of Endocrine Disruption by Environmental Chemicals. Gretchen Kruse, Idaho Department of Fish and Game, Coeur d Alene, ID. Reproduction in fish relies on proper production and function of hormones in the neoroendocrine system. These functions can be disrupted by actions of exogenous environmental chemicals that bind to receptor cites or mimic the actions of hormones. Measuring plasma steroid levels in fish provides a means for monitoring functions in the neuroendocrine system. Due to limited reproduction, the Kootenai River white sturgeon (*Acipenser transmontanus*) was listed as an endangered species in 1994. One of the potential limiting factors in this population is environmental contamination due to basin-wide municipal, industrial, mining and agricultural operations. These contaminants have the potential to disrupt normal functions in the endocrine system and in effect, limit reproduction. During March 1998, thirty-nine blood samples were drawn from Kootenai River white sturgeon at various levels of sexual development. The blood samples were tested for testosterone, 11-ketotestosterone and estradiol levels as biomarkers to determine potential effects from environmental contamination.

PWA212 Assessment of the Potential Male Reproductive Toxicity of the Gasoline Additive Methyl tert-Butyl Ether Utilizing a Testosterone Biomarker. Billitti, J.E., Faulkner B.C.*, Lasley, B.L., and Wilson, B.W., University of California, Davis. One of the primary concerns regarding the gasoline additive methyl tert-butyl ether (MtBE) is contamination in drinking water and the potential impact of exposure on reproductive health. Our research group has recently developed and validated a non-invasive assay for testosterone using ether extracts of feces to assess the reproductive state in male mice exposed to MtBE. A pilot study was conducted in which Swiss-Albino mice were gavaged with three dose levels of MtBE (400 mg/kg, 1000 mg/kg, and 2000 mg/kg) and fecal testosterone levels, end point serum testosterone, animal and testes weights were examined. The results of this preliminary study did not demonstrate significant decreases in any of the parameters examined. Histopathology of the testes of exposed animals is currently underway. Future experiments will evaluate the reproductive toxicity of environmental degradation products of MtBE. (Supported by UC Toxic Substance and Teaching Program; SB521)

PWA213 The Response of Certain Biomarkers in PCB and Chlordane Contaminated Paddlefish (*Polyodon spathula*), from the Ohio River Basin. Gundersen, D.T., Krahling, M., Bauermeister, M.*, Sanders, K., The University of Southern Indiana, Evansville, IN; Mims, S., Aquaculture Research Center, Frankfort, KY; Millar, J., U.S. Fish and Wildlife Service, Rock Island Ill. Ohio River paddlefish populations may be struggling due to habitat loss, and exposure to chlorinated hydrocarbons. This study determined PCB and chlordane concentrations in paddlefish gonads. Plasma sex steroids, hepatic microsomal ethoxyresorufin O-deethylase (EROD) activity, and percent hatch were also looked at. Thirty-one paddlefish were collected from the Ohio River, and 20 paddlefish were collected from the Cumberland River. Chlordane (0.49 ± 0.08 ppm) and PCB (0.83 ± 0.17 ppm) concentrations in Ohio River paddlefish ovaries were significantly higher than chlordane (0.07 ± 0.01 ppm) and PCB (0.06 ± 0.04 ppm) concentrations in Cumberland River paddlefish ovaries. Higher chlordane ($2.10\pm.27$ ppm) and PCB (4.01 ± 0.81 ppm) concentrations were also detected in the testes of Ohio River paddlefish compared to chlordane (0.30 ± 0.07 ppm) and PCB (0.34 ± 0.08 ppm) concentrations in Cumberland River paddlefish testes. PCB congener specific analysis indicated that the tetra-, penta- and hexachlorobiphenyls were the most prevalent PCBs in gonad samples. Male paddlefish collected from the upper Ohio River had significantly lower plasma testosterone levels (9.7 ± 1.6 ng/mL) than males (16.9 ± 1.5 ng/mL) and females (20.93 ± 0.57 ng/mL) collected from the lower river and the Cumberland River (males = 19.0 ± 1.3 ng/mL; females = 16.9 ± 1.5 ng/mL). None of the fish collected had measurable EROD activity. Percent hatch was greater than 86% in 6 female paddlefish that were collected and induced to spawn. The lack of EROD activity in paddlefish indicates that this may not be a useful biomarker. The low plasma testosterone levels seen in upper Ohio River paddlefish may be due to the elevated PCB/chlordane concentrations seen in these fish. Percent hatch did not appear to be affected by these contaminants but further studies are warranted on paddlefish offspring survival.

PWA214 Effects of 2,3,7,8-Tetrachlorodibenzo-p-Dioxin on Immune Functions and CYP450 in Adult Male Deer Mice (Peromyscus maniculatus). Liu, J.*, Peden-Adams, M. M., Kunteson, S., Dancik, J., Bodine, A. B., Department of Environmental Toxicology, Clemson University, Pendleton, SC.
2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) causes a broad range of immunotoxic effects including immunosuppression of nonspecific as well as specific cell-mediated and humoral immunity. TCDD administration to experimental animals also results in CYP450 induction. The objectives of this study were to determine the effects of TCDD on immune function and CYP450 in a non-traditional mammalian species. In this study, 7-8 week old male deer mice (Peromyscus maniculatus) were dosed via ip injection every other day for 12 days. Five concentrations of TCDD (0.3, 1, 3, 10, and 30 mg/kg) and a corn oil vehicle (control) were used for the dose response study. Immune function was

assessed using T- and B- lymphocyte blastogenesis, plaque forming cell assay (PFC) and macrophage phagocytosis to determine potential effects on cell-mediate immunity, humoral immunity, antibody production, and non-specific immunity. T- and B- cell proliferation along with the PFC assay was suppressed when compared with control. Macrophage phagocytosis, however, was stimulated. EROD, as expected, was induced when compared with control. These results indicate that at levels as low as 1 mg TCDD/kg humoral immune function is significantly suppressed in this non-traditional mammalian species.

PWA215 Fluoranthene and 1,2-Benzanthracene Exposure in Adult Male Deer Mice (*Peromyscus maniculatus*) Results in Alteration of Immune Function. Liu, J.*, Peden-Adams, M. M., Kunteson, S., Dancik, J., Bodine, A. B., Department of Environmental Toxicology, Clemson University, Pendleton, SC. Polynuclear aromatic hydrocarbons (PAHs) are among the most common chemicals found in uncontrolled waste site. These chemicals produce a number of toxic and biochemical responses in most species. However, immune function alterations are often overlooked. The objectives of this study were to determine the effects of two PAHs, fluoranthene and 1,2-benzanthracene, on immune function in a non-traditional mammalian species. In this study, adult male deer mice (*Peromyscus maniculatus*) were dosed with either fluoranthene or 1,2-benzanthracene every other day for 12 days. Five concentrations of these chemicals (0.3, 1, 3, 10, and 30 mg/kg) and a corn oil vehicle (control) were used. Immune function was assessed using both T- and B- cell proliferation, antibody plaque forming cell response (PFC) and macrophage phagocytosis to determine potential effects on cell-mediate immunity, humoral immunity, antibody production, and non-specific immunity. 1,2-benzanthracene exposure resulted in a bell-shaped dose-response for the T- and B- cell proliferation as well as the macrophage phagocytosis. PFC assay exhibited a dose-responsive suppression for both chemicals. There was no obviously effects of FAE on T- and B- cell proliferation or on macrophage phagocytosis.

PWA216 Alterations In Immune Function Of Adult Male Deer Mice Following Exposure To Either 7,12-Dimethylbenzanthracene Or Benz[e]acephenanthrylene. Peden-Adams, M.M.*, Liu J., Dancik, J., Knuteson, S., Bodine, A.B., Department of Environmental Toxicology, Clemson University. The realization that wildlife immunity can be affected by environmental contaminants has stimulated the use of immunotoxicity in the field of environmental toxicology with hopes of assessing the effects of contaminates on wildlife health as a basis for environmental risk assessment. The use of immunotoxicology as a biomarker of environmental exposure has great potential once it is validated in non-traditional sentinel species. The objective of this study was to determine the effects of two PAHs on immune function in a nontraditional murine species. PAHs are widespread environmental contaminates which can be found on many uncontrolled hazardous waste sites. Adult male deer mice were exposed every other day for a period of twelve days to the chemicals (0.3,1,3,10, and 30 mg/kg) or the corn oil vehicle. Immune endpoints assesed were lymphoproliferation, macrophage phagocytosis, and the antibody plaque forming cell response. 7, 12-dimethylbenz[a]anthracene (DMBA) exhibited a bell shaped dose-response in both T- and B-cell proliferation, while benz[e]acephenanthrylene demonstrated a biphasic response. Both PAHs, however, caused a dose-responsive suppression of the antibody plaque forming cell assay. DMBA exposure also resulted in a dose-responsive suppression of phagocytic activity, while benz[e]acephenanthrylene affected a biphasic response. It is evident that each PAH has its own specific effect on the components of the immune system; therefore, the model PAHs (ie DMBA) can not accurately predict the effect of other PAHs.

PWA217 Altered Immune Functions in Fish Predict Toxicological Hazards Associated with Exposure to Chemical Mixtures. Raymond, A.D.*, Li, Y., Zelikoff, J.T. Dept. of Env. Medicine, New York University School of Medicine. A battery of immune assays, originally developed in rodents, has been adapted for use in Japanese medaka to evaluate toxicological hazards associated with contaminated aquatic environments. Fish were exposed for either 3, 7, or 14 d in the laboratory to groundwater collected from an organic- and metal-contaminated groundwater aquifer at Aberdeen Proving Grounds (MD). Following exposure to either 10, 50, or 100% groundwater, fish were sacrificed and their kidneys/spleens used to provide cells for immune assays; blood/serum was recovered for differential counts and measurements of hematocrit, leukocrit, and immunoglobulin (Ig) levels. Exposure for 3 d to undiluted groundwater production was enhanced. Formation of hydrogen peroxide (H_2O_2), hematocrit, and kidney cell numbers were also reduced, but only in fish exposed to 50% groundwater. Following a 7 d exposure to undiluted groundwater, proliferation by splenic lymphocytes and H_2O_2 and O_2^- production was significantly reduced (compared to control); plasma Ig levels were elevated compared to the control and 3 d exposure group. Exposure for 14 d reduced survival in the 100% exposure group by ~90%; immune organ and body weights decreased and increased, respectively in both the 10% and 50% exposure groups. Plasma Ig levels were slightly increased (compared to control) after 14 d in both the 10 and 50% exposure groups; O_2^- and H_2O_2 production were also altered in these treatment groups. These studies demonstrated the applicability of immune assays performed in laboratory fish to predict the toxicological hazards associated with mixtures of contaminants found in polluted aquatic environments. Supported by U.S. Army Center for Environmental Health Research DAMD-17-93-C-3059 and Army Augmentation Award

PWA218 The Immune Response of Fish: a Sensitive Indicator of Permethrin Toxicity. Zelikoff*, J.T., Li, Y., Carlson, E., and Raymond, A. New York University School of Medicine, New York, NY. Permethrin, used as an insect repellent by some humans, is a pharmacologically active synthetic compound noted for its insecticidal properties. While effects of pyrethroids upon the nervous system have been fairly well-studied, little information is available concerning effects of this class of compounds on the immune response. Considering the potential for human exposure, a better understanding of the health risks associated with short-term low-dose permethrin exposure is critical. Japanese medaka (Oryzias latipes) were exposed in the water to alcohol-solubilized permethrin for 48 hr at nominal concentrations between 0.01 and 15 ppb; the 48 hr LC50 was ~0.26 ppb. Results of these studies demonstrated that while exposure to permethrin had no significant effects upon thymus histology, permethrin concentrations „ 0.05 ppb reduced (compared to unexposed controls) plasma immunoglobulin levels and/or kidney and spleen cellularity in a dose-dependent manner. Alternatively, exposure to permethrin increased the proliferative response of splenic lymphocytes, as well as melanomacrophage number/size compared to controls; intracellular and extracellular superoxide production by kidney phagocytes was also altered by exposure to permethrin. Moreover, exposure of fish to permethrin at a concentration well-below the lC50 value reduced host resistance against an infectious bacterial pathogen. These findings demonstrate the usefulness of immune assays developed in rodents to predict the immunotoxicity of permethrin in a fish model, as well as the ability of permethrin to alter host immunocompetence. Supported by U.S. Army Center for Environmental Health Research DAMD-17-93C-3059.

PWA219 Evaluation of the immunotoxicity of coplanar PCBs and butyltins exposure in human and marine mammals by measuring mitogen-induced responses of peripheral blood mononuclear cells (PBMC). Nakata, H.*, Kanoh, M., Sakakibara, A, Ehime University, Matsuyama, Ehime, Japan; Kudo, S., Marine World Uminonakamichi, Fukuoka, Fukuoka, Japan; Miyazaki, N. University of Tokyo, Otsuchi, Iwate, Japan; Tanabe, S. and Asano, Y., Ehime University, Matsuyama, Ehime, Japan. To investigate immunotoxicity of coplanar PCBs (IUPAC 77, 126 and 169) and butyltin compounds (tributyltin; TBT, dibutyltin; DBT, monobutyltin; MBT) in marine mammal, proliferative responses of PBMC were measured after stimulation with different concentrations of mitogens such as concanavaline A (Con A), phytohaemagglutinin (PHA) and pokeweed mitogen (PWM), with contaminants. The optimal mitogen concentrations were examined in marine mammals, and the responses to Con A, PHA and PWM were maximal at 10 μg/ml, 5 μg/ml and 1 μg/ml, respectively. Maximum activation were found during 4th day of culture periods. Con A responses were inhibited apparently with the dose of 300 nM of TBT and 330 nM of DBT in cetaceans, while MBT showed little inhibition even with a concentration of 3,300 nM. In case of human, TBT and DBT also showed the inhibition of response in the concentrations of 30 and 330 nM, respectively, but no inhibition was found in MBT exposure. This suggests that TBT and DBT are more immunotoxic than MBT. PCBs did not markedly affect the response at the concentration tested (2.7 pM34 nM) in human. Since TBT and DBT levels showing the inhibition in this study are close to those found in the marine mammals inhabiting coastal water in Japan and Northern America, some populations of coastal cetaceans may be at risk to immunotoxicity.

PWA220 Linkage of Histopathology to Body Burden and Condition Index in Feral Asian clam (*Potamocorbula amurensis*) exposed to metallic contaminants. Clark S.L.* and D.E. Hinton, University of California - Davis, Davis, CA. Our biomarker toolkit for field studies is heavily weighted toward markers of exposure but markers of deleterious effect are needed if we are to determine risks arising from exposure. Previous work, performed over multiple years, has determined levels of metallic contaminants in clams and related this to individual level effects (condition index). This approach has demonstrated populations with poor condition index along a contaminant gradient in the upper San Francisco Bay. This study was designed to evaluate histologic alterations within individuals of affected clam populations and to determine whether degree of alteration was related to contaminants and condition indices. Monthly (weather permitting) collections were made form four sites along the above gradient. Visceral masses were dissected free, fixed, dehydrated and embedded in paraffin. Following sectioning (6-8 µm) and mounting on glass slides, sections were stained with hematoxylin and eosin for survey of all organs and tissues. Digestive diverticulum, gonad (both sexes) and gill were sites of histopathologic alteration. Prevalences of lesions as well as temporal and spatial relationships of affected clams were established. Interestingly, clams from the station with the highest body burden, lowest condition index, and heterogeneous gonadal development were the individuals with the greatest prevalence of gonadal lesions. Supported in part by the USEPA - UC Davis Center for Ecological Health Research #819658 and by grant # 823297 from the USEPA. SC is a graduate trainee in the UC systemwide Toxic Substances Research and Teaching Program's lead campus program in ecotoxicology.

PWA221 A sublethal toxicity evaluation of pollutants for fish using hematological parameters. Kakuno A.* and Koyama J., National Research Institute Fisheries Science, Nagai, Yokosuka, Kanagawa, 238-0316, Japan. Most of no observed effect concentrations (NOECs) of pollutants have been determined by survival rate or growth rate. Some hematological parameters are affected at lower concentrations of pollutants. Therefore, NOECs of some pollutants for popular cultured marine fish in Japan, red sea bream *Pagrus major* (BW 70-280g) were determined by growth rate and following hematological parameters, red cell count, hematocrit (Ht), hemoglobin (Hb), plasma glucose lactate concentrations, lipid peroxidation and Glutathione concentration in red cells. Red sea bream which is popular marine fish in Japan, were exposed to sublethal concentrations of triphenyltin chloride (TPTC; 0.12, 1.05 and 2.93 TPT mg/l), bis-trybuthyltin oxide (TBTO; 0.031, 0.112, 0.55 and 1.98 TBT mg/l), cadmium (Cd; 0.01, 0.06 and 0.37 Cd mg/l) and naphthalene (0.06, 0.21, 0.8 and 2.42 mg/l) for 8 weeks. After exposure to TBTO, NOEC, 0.52 TBT mg/l, was determined by Ht and MCHC (mean corpuscular hemoglobin concentration). NOEC of TPTC, 1.05 TPT mg/l, was determined by red cell count and plasma lactate concentration. NOEC of Cd, 0.06 Cd mg/l. was determined by Ht and lipid peroxidation in red cells. NOEC of naphthalene, 0.06 mg/l, was determined by red cell count, Ht, Hb and plasma glucose concentration. These NOECs were lower than NOECs determined by growth rate. This study suggested that the effects of these pollutants to fish can be detected sensitively by red cell count or Ht rather than other hematological parameters and growth rate.

PWA222 Study of LF25 Polypeptide as Biomarker to Heavy Metal Exposition in *Limnoperna fortunei*. Belaich, M.; Oliver, C. & Porta, A.*. Laboratorio de Ecotoxicología y Química Ambiental, Departamento de Ciencia y Tecnología, Universidad Nacional de Quilmes. Utilization of biological response to watch contamination extent allows obtaining information that it can not acquired by chemical traditional methods. In this matter, it is necessary to have sentinel organisms and biomarkers to detect the exposition to xenobiotics as heavy metals in sensible way. Bivalve mussels meet the requisites to be used as biomonitoring organisms. Its sedentary and alimentary habits (feeding filter organisms) allow accumulating fast and efficiently xenobiotics. New imported specie is being evaluated. *Limmoperna fortunei* is a bivalve from Asia like *Corbicula fluminea*, and it was carried to Rio de la Plata in similar form. In particular, it is very important to analyze the utility of the considered biomarkers and the effect of the seasonal variables (as temperature, salinity, pH, and organic matter contents changes) on this response. In this work we evaluate the response of polypeptide LF25 induced in *Limnoperna f.* exposed to Cd (II) and Cu (II) from the biomarker of contamination point of view. Mussels were sampled from the Rio de la Plata in a non-polluted zone. They were acclimated one week in synthetic water previously to heavy metal stress. The animals were exposed for 5 days. Experience were repeated using the maximum and minimum local values of temperatures, salinity, and dissolved humic substances concentration. From previous experiences, entire animal was taken. The tissue was homogenized with buffer Tris-Sucrose and the homogenate, with the same protein concentration (Bradford method), was analyzed by electrophoresis using the Tris-Tricine system. Polypeptides were detected by Coomassie Blue staining method. Also were assayed the response using HPLC/ODS.

PWA223 Spatial and Temporal Profiles of Stress Protein (HSP70) and Metallothionein in Asian Clam (*Potamocorbula amurensis*) in Northern San Francisco Bay. Werner, I.*, Clark, S.L, Kaufman, R.C., and Hinton, D.E., University of California, Davis, CA. Multiyear investigations by United States Geological survey have revealed populations of Asian clam (*P. amurensis*) with reduced condition indices along a well established metal contamination gradient in Northern San Francisco Bay. Our study seeks to determine whether biomarker responses such as stress protein hsp70 levels and concentrations of sulfhydryl-rich, *metallothionein-like* proteins in *P. amurensis* can be correlated with exposure to anthropogenic stressor(s), and/or are related to histopathologic markers and the individual's condition. Here we present our results on stress proteins (hsp70) and metallothionein. Stress proteins are induced by a variety of stressors, which either damage cellular proteins directly or cause cells to synthesize aberrant proteins. Low molecular weight, sulfhydryl-rich proteins and polypeptides, i.e. metallothioneins and phytochelatins, have been reported to be important in both metal homeostasis and detoxification. Clams were sampled monthly from 4 stations in Northern San Francisco Bay over the period of 2 years (June '96 - June '98). Stress proteins are analyzed by western blotting using monoclonal antibodies. Levels of metallothionein-like proteins are determined using SDS-PAGE electrophoresis with monobromobimane as a fluorescent probe specific for free sulfhydryl moities.

PWA224 Evidence of Metalloproteins in Copepoda Harpacticoida *Trigriopus brevicornis* (Muller) Exposed to Mercury and Cadmium. Barka,S., Institut Oceanographique-Paris, France; Pavilon, J.F., Institut Oceanographique-Paris, France; Amiard-Triquet, C., Faculte' de Pharmacie, Service d'Ecotoxicologie, Nantes, France. Although metalloproteins have been found in different organisms, only a little is known about the large group of Copepods which represents more than 70% of oceans planktonic biomass. Copepods were collected along the french North Atlantic coast near the Loire Estuary (Le Croisic). *Tigriopus brevicornis* ovigerous females were exposed to different concentrations of non-essential metals: inorganic Hg and Cd. The isolated metalloproteins were quantified with Differential Pulse Polarography. A dose-response and a time-course experiments were performed. Metalloproteins were significantly induced in a dose-dependent way in response to both waterborne $CdCl_2$ and $HgCl_2$. A significant correlation was found between metals and induced metalloproteins.

PWA225 Metallothionein as a biomarker for trace metal exposure and effects in the fresh water oligochaete, *Tubifex tubifex.* P. Gillis*, University of Waterloo, Waterloo, Ont., T. B. Reynoldson, National Water Research Institute, Burlington, Ont., D.G. Dixon, University of Waterloo, Waterloo, Ont., and L. C. Diener, University of Waterloo, Waterloo, Ont. The freshwater oligochaete, *Tubifex tubifex* was used in the development of a biomarker to quantify metal exposure in benthic invertebrates. Oligochaetes were chosen as a representative benthic invertebrate because they are in constant contact with the sediment both through bodily contact and ingestion of the sediment. Laboratory cultured *T. tubifex* were exposed to trace metals in artificially contaminated field sediment. Following exposure, metallothionein concentrations were measured using a mercury saturation assay based on Dutton et al., (1993). Metallothionein concentrations in the worm tissues were found to exhibit a dose response relationship with metal exposure. Reproductive output of the exposed worms was determined in order to determine if elevated metallothionein concentrations in the tissues of *T. tubifex* correspond to an ecologically relevant endpoint. Results from 28 day whole sediment bioassays indicate that reproductive output in *T. tubifex* is negatively affected by metal exposure and is inversely related to tissue metallothionein concentration. A time series experiment was conducted in order to determine the time to induction of metallothionein synthesis. Metallothionein concentrations were monitored in *T. tubifex* exposed to metal contaminated sediments for a period of 0 to 1000 hours. Induction of metallothionein synthesis in *T. tubifex* appears to be rapid with a significant increase ($p<0.05$) after eight hours of exposure to metal contaminated sediments and reaching a maximum after 24 hours exposure.

PWA227 Accumulation of Heavy Metals and Metal-Binding Stress Proteins as Biomarkers of Potential Toxicity to Cichlid Fishes from the Rio Negro, Brazil. Howard, C.L.*, Paulos, P., University of Houston – Clear Lake, Houston, TX; Quast, W.D., Parametrix, Inc., Houston TX. Although mineral extraction, deforestation, agriculture and industrial development and their associated pollution impacts are of increasing concern to ecological health of neotropical rain forests, there has been very little work investigating the ecotoxicological implications of these activities. In fact, no such studies have been conducted on the Rio Negro, a sensitive, naturally acidic habitat and the largest tributary of the Amazon River, and which is presently subjected to increasing gold mining, deforestation and urbanization. The purpose of our study was to analyze the heavy metal content of two fish species of dietary importance and correlate any metal accumulations in these fish with data from sediment and water analyses. We collected peacock bass (*Cichla temensis*) and black piranha (*Serrasalmus* sp), as well as water and sediment samples, from 6 sites along the Rio Negro between Manaus and Barcelos, Amazonas, Brazil during January 1998. Fourteen water chemistry parameters were measured on site, and heavy metals in water and sediment were analyzed by ICP or graphite furnace upon return to Houston. Samples of fish muscle and liver tissue were dissected out and frozen immediately in liquid N for transport to the lab, where they were digested and analyzed for Hg, Pb, Cd, Cu, Ni, Zn and Cr. Profiles of liver tissue metal-binding proteins were established for each species and were correlated with heavy metal content of the fishes. The results of this study may be considered baseline and will be compared to further work being completed during the summer of 1998 on fish collected from heavily polluted sites around Manaus.

PWA228 Effects of Parathion, Fenitrothion and Paraoxon on Glutathione S-transferases, Cholinesterases and Carboxylesterases in *Corbicula fluminea* of the Rio de la Plata. Basack, S.B.*, Oneto, M.L., Fuchs, J.S., Wood, E.J., Kesten, E.M. Facultad de Ciencias Exactas y Naturales, Universidad de Buenos Aires, Argentina. The agricultural chemicals represent a significant source of pollution in aquatic environments. There is little information about the responses of freshwater bivalves to organophosphorus pesticides (OPs). Our aim was to assess the effects of parathion, fenitrothion and paraoxon on cholinesterases (ChEs), glutathione S-transferases (GST) and carboxylesterases (CaE) of *Corbicula fluminea*. These bivalves were collected during March and April 1998 from a non polluted zone of the coast of the Rio de la Plata, and they were acclimated in the laboratory to test conditions. The bioassays were performed one week after collection. The organisms were exposed during 96 h to pesticides concentrations from 0 to 2000 µg/L. The whole soft tissue was homogenized and centrifuged in order to obtain the 10000 x *g* pellet and supernatant. These fractions were used for the enzymatic studies. The ChEs and CaE activities were assayed by the method of Ellman (1961) and the GST activities were measured according to Habig (1974). After a 96 h exposure period to the OPs, no mortality occurred in controls and exposed bivalves. CaE activities were depressed at all the concentrations assayed. The pellet ChE was significantly inhibited at concentrations as low as 20 µg/L of parathion, 50 µg/L of fenitrothion and 10 µg/L of paraoxon. Similar results were observed for the supernatant ChE except for paraoxon which was able to inhibit this enzyme at an even lower concentration (2 µg/L). The activity of GST was increased in organisms exposed to 2000 µg/L of fenitrothion and 150 – 500 µg/L of paraoxon.

PWA229 Effects of Pollutant Discharge on Populations of Marine Intertidal Copepod *Tigriopus brevicornis* (Copepoda) in the Field. Forget J.*, Institut Oceanographique, Paris, France , Bocquene' G., Ifremer, Nantes, France. During on year, field studies were carried out at five stations on the west coast of France. The ((Vilaine)) estuary was chosen per two essential grounds, on one hand the tank basin spreads third of the Brittany, of which the vocation is essentially agricultural and is greatly contaminated by the pesticides, on the other hand *Tigriopus* is present in the intertidal flat of the estuary and permits the exploitation like species sentry. The levels of AChE detected in copepods showed indications of decreasing AChE activity with increasing presence of pollutant. The highest AChE activity was found at the far site from the estuary and a lowest activity was observed to the station pleased her to the interior of the estuary. This last station is the most exposed to the contributions of pesticides according to the chemicals analyses. A clear dose-response relationship was observed, the inhibition of AChE activity in *Tigriopus brevicornis* could be considered to be a useful sublethal indicator of marine environmental pollution. and a good candidate for pollution monitoring.

PWA230 The Effect of Dieldrin on Biogenic Amine Levels and Torpor in the Deer Mouse, *Peromyscus Maniculatus*. Montie, E.W.*, Pivorun, E.B., McAuley, A.P., Clemson University, Clemson, SC; Hooper, M.J., Texas Tech University, Lubbock, TX. From the 1940s to the late 1970s, the Rocky Mountain Arsenal (RMA) was a site for the manufacture of dieldrin. High concentrations pollute the soil posing potential threats to wildlife. Studies indicate that dieldrin increases metabolism and may disturb brain biogenic amines. Since biogenic amines are regulatory neurotransmitters controlling an animal's metabolic shift into torpor, exposure to dieldrin may disrupt the ability to cope with harsh winter conditions. The experiment used wild deer mice, *Peromyscus maniculatus*, as a model. In Tier I, a four week preliminary dosing study with four levels (.01, 0.1, 1.0 and 10 ppm dieldrin in diet) was carried out. In Tier 2, an eight week study with two dose levels (1 and 10ppm in diet) was performed. At the end of four weeks, torpor was induced and monitored daily. For both tiers, endpoints included brain residue and biogenic amine levels, and cytochrome P450 activity. Tiers 1 and 2 showed elevated EROD induction at 10 ppm. Tier 1 levels went from 241 (control) to 444.9 pmoles ethoxyresorufin/min/mg protein (10ppm). Tier 2 levels went from 297 (control) to 567 pmoles ethoxyresorufin/min/mg protein (10ppm). In Tier 2, preliminary data show the 10 ppm dose group to have a 1.7 fold increase in DOPAC levels versus the control group, indicating an increase in dopamine release and metabolism. During tier II, one mouse died at each dose, indicating a possible torpor disturbance. Preliminary data show an increase in torpor duration at the 10ppm dose. Sponsored by USFWS RMANWR and NIH ES04696.

PWA231 The imposex response in *Morula granulata* as bioindicator of tributyltin (TBT) contamination in the Dampier Archipelago, Western Australia. Reitsema, T.J, Curtin University, Perth, Western Australia. This field survey was undertaken to examine the use of the neogastropod whelk *Morula granulata* as bioindicator of TBT contamination in the Dampier Archipelago. TBT in this area originates from commercial vessels using the Port of Dampier that are >25 m in length and exempt from legislation controlling the use of TBT in antifouling paints. Concern has arisen regarding the effect that TBT has, both on non-target organisms, and possible connections of this endocrine disruptor to adverse trends in human health. In July 1997, a field survey was undertaken; 100 *Morula granulata* were taken from 18 sites of varying vessel activity throughout the Archipelago, and examined for imposex. The level of TBT contamination in the Archipelago was low; ranging from <0.3 to 25 ng Sn l^{-1} in water, <0.3 to 80 ng Sn g^{-1} in oyster tissue (*Saccostrea cucullata* - prey of *M. granulata*), and <0.3 to 33 ng Sn g^{-1} in whelks. Percentage of imposex in female *Morula granulata* ranged from 0 to 57%, and was strongly correlated with vessel activity ($r = 0.85$). The only significant (at $\alpha= 0.05$) relationship of imposex with butyltin concentrations was with TBT in oysters (r =

0.826, p = 0.000), and dibutyltin (DBT) in whelks (r = 0.783, p = 0.000). It is postulated that this is because of interspecies variation in ability to reduce TBT to DBT and MBT (monobutyltin), as well as induction of imposex in juveniles, so that imposex could be a reflection of prior contamination. *M. granulata* is a suitable bioindicator of TBT contamination in the Dampier Archipelago, which could be used throughout the Indo-Pacific given its abundance in the region.

PWA232 Chironomid Mentum Deformities as a Biomarker of Oil Sands Process Water Toxicity in Wetlands of Northern Alberta. Whelly, M.P., Ciborowski, J.J.H.*. University of Windsor, Windsor, ON. The effects of oil sands process water (OSPW) on aquatic invertebrates in wetlands near Fort McMurray, AB, was assessed using mouthpart deformities in midges (Diptera: Chironomidae). This teratogenicity biomarker has been associated with reduced emergence success. OSPW is saline, and contains trace metals, PAHs and naphthenic acids. Taxa richness and invertebrate abundance were also measured. Fifteen wetland sites were sampled, three of which were OSPW affected. Principal components analysis and cluster analysis of environmental characteristics (water pH, conductivity, salinity, dissolved oxygen, sediment particle size distribution and organic content) were used to identify pairs of environmentally similar wetlands that differed mainly in exposure to or absence of OSPW. Dip net and Ekman grab sampling were performed to assess invertebrate community composition. Additionally, 250 large, red chironomid larvae were hand-collected for deformities analysis. Taxa richness was moderately but not significantly lower at OSPW affected wetlands compared to corresponding reference sites (1-tailed paired-comparison t-test, p=0.07). There was no difference in abundances (1-tailed paired-comparison t-test, p>0.1). The incidence of mentum deformities (only missing or additional teeth considered) in field-collected chironomids from the three OSPW affected and three corresponding reference wetlands ranged from 1.4 - 3.7 % and 0.0 - 2.8 %, respectively. There was no difference in levels of deformities among sites (G-statistic test of homogeneity = 5.0051, p>0.5) or between paired reference and OSPW affected wetlands (G-statistic test of independence, p>0.1 in all 3 cases). The suspected teratogens in OSPW (trace metals, PAHs) may not be bioavailable in these highly humic wetlands.

PWA233 Analysis of Monosaccharides in *Chironomus riparius* Using Gel Electrophoresis. Bentivegna, C.S.* and Davitt, J., Seton Hall University, South Orange, NJ. The composition and quantity of monosaccharides were evaluated as a biomarker for physiological stress in *Chironomus riparius* using the new technique of carbohydrate gel electrophoresis. Little work has been published on this technique, so a number of tests were run to try to standardize the procedure. Groups of 6-10 chironomids were fed or starved from 24 to 120 h.. Test chambers consisted of 60 g of acid washed sand and 250 ml of carbon filtered water. Fed organisms received 3 drops of ground fish food pellets (0.1 g/ml) daily. Monosaccharide analyses were performed on whole tissues. Samples were prepared by tissue homogenization, acetonitrile precipitation of proteins, chloroform extraction of lipids, and hydrolysis of carbohydrates using trifluoroacetic acid. Monosaccharides were then labeled using fluorophore 2-aminoacridone, separated on a monosaccharide composition gel (BIO-RAD) and visualized on a UV illuminator. Results showed a progressive reduction in monosaccharide levels starting at 48 h. Detection limits were 0.01 mg/ml glucose and 0.05 mg/ml for allose. There were three predominant bands in the fed groups. One co-migrated with glucose but the other two have not yet been identified. Known standards that were tested included N-acetylgalactosamine, N-acetylglucosamine, allose, altrose, fructose, fucose, galactose, glucose, mannose, sorbitol, and talose. Allose (0.2 mg/ml) was chosen as an internal standard as it did not co-migrate with any of the predominant bands. Advantages of this technique included the low detection limit, quantification by densitometry, and detection of unknowns. Disadvantages included cost and the ability to detect monosaccharides only.

PWA234 Evaluating Biochemical Biomarkers as Potential Environmental Monitoring and Protection Tools. McLoughlin, N.J.*; Maltby, L.; Wood, R., University of Sheffield, Sheffield, U.K.; Yin, D.; Yu, H., Nanjing University, Nanjing, China. The application of biomarkers is receiving considerable attention as a potentially powerful set of diagnostic tools for assessing exposure to toxicants. Measurable changes at the molecular and biochemical level are likely to occur at an earlier stage of exposure than changes at higher levels of biological organisation (e.g. reproduction). Biomarkers as diagnostic tools could be used to establish the presence of specific contaminants together with the chemical fate and transport of a pollutant. Our limited understanding of biomarker responses is significantly hindering their application. The objective of this study were to evaluate two established biochemical biomarkers for use as freshwater diagnostic tools. The biomarkers chosen were the inhibition of cholinesterases and the induction of glutathione-s-transferase. Biomarker methodologies were optimised for use with the amphipod, *Gammarus pulex*. For each assay the limit of quantitation was determined and power analysis conducted to assess appropriate replication numbers. To assess the specificity of the biomarkers animals were exposed to five major classes of chemicals; heavy metals, OPs, pyrethroids, OCs, surfactants, and enzyme activities were determined. Additionally, to extend and validate the utility of biomarkers, animals were deployed *in situ* at 10 sites of established point source pollution and 1 site receiving pesticide spray drift. Major contaminant types associated with the selected discharges included: heavy metals; surfactants; pesticides; disinfectants; organic enrichment and chlorine. In this presentation, the authors give a synthesis of the both the laboratory and field studies and discuss the implications of biomarker specificity and sample variation in relation to the potential use of biomarkers as environmental monitoring and protection tools.

PWA235 Bioindicator Studies with Black Flies: Acute Toxicity of Chlorpyrifos to Instar Groups of *Simulium Vittatum* Is-7 and *Simulium Vittatum* IIIL-1. Hyder, A.* and Noblet, R., Department of Environmental Toxicology, Clemson University, Clemson, SC. Larval black flies are presently not on the United States Environmental Protection Agency's list of standard organisms used in toxicity testing. *Simulium viattatum* IS-7 and *S. vittatum* IIIL-1, two sibling species of black flies, are ecologically diverse organisms that inhabit well-aerated, fairly pristine areas and highly organically loaded areas, respectively. A colony of *S. vittatum* IS-7 larvae and S *vittatum* IIIL-1 eggs reared to larvae in the laboratory were used to conduct acute toxicity tests at different developmental stages. Chlorpyrifos, a widely used organophosphate, was the toxicant to which the organisms were exposed for 24 hours. Tests were conducted in a bioassay system designed to simulate a stream exposure. Preliminary results suggest a difference in the mortalities of these two species during different instars to chlorpyrifos over the 24 hour exposure period.

PWA236 Suitability of *Hygrohypnum ochraceum* (Bryophyta) as an Indicator of Inorganic Pollutants in Streams and Rivers of North America: Laboratory Studies. Montaño, A.M., U.S. Bureau of Reclamation, Denver, CO. Aquatic bryophyte tissues were exposed to cadmium, zinc and selenium to assess their 10-day bioaccumulation potential and to determine physiological stress-response effects under laboratory conditions. Moss tufts of *Hygrohypnum ochraceum* (Turn. ex Wils.) Loeske, Moosfl. Harz. were collected from Nate Creek Ditch, Colorado USA. Mosses were acclimated to laboratory conditions and were then exposed to Cd, Zn and Se in independently run experiments at specified concentrations for the bioaccumulation portion of this study. Metal treatments for each experiment were performed using a static-renewal method. Physiological stress-response measurements were performed at the end of each bioaccumulation experiment by measurement of chlorophyll-to-phaeophytin ratios (OD665/OD665$_a$). All experimental metals were accumulated in *H. ochraceum* tissues. Average daily uptake was 69.8% for Cd, 72.2% for Zn, and 10.6% for Se of their initial concentrations. Physiological stress-response measurements showed no significant differences between control- and metal-treated samples. *H. ochraceum* appears to successfully accumulate metals under certain environmental conditions. These environmental factors also seem to effect determination of physiological stress-response measurements.

PWA237 Biomarkers for Contaminant-Associated Immunosuppression in Colonial Waterbirds of the Great Lakes. Grasman, K.A.*, Wright State University, Dayton, OH; Fox, G.A., Canadian Wildlife Service, Hull, PQ. Persistent organochlorines, especially polychlorinated biphenyls (PCBs) and 2,3,7,8-tetrachlorodibenzo-*p*-dioxin, are powerful immunosuppressants in birds and mammals. Our previous investigations have found associations between organochlorines and suppression of T cell-mediated immunity in herring gull (*Larus argentatus*) and Caspian tern (*Sterna caspia*) chicks at highly contaminated sites in the Great Lakes. This study further investigated these immunotoxic effects, employing additional biomarkers and assessing organochlorine concentrations in individual birds. The development of immunological organs was assessed in pipping herring gull embryos and four week old chicks. *In vivo* immune function tests were conducted on three to four week old gulls and terns. Herring gull embryos at two highly contaminated sites (Saginaw Bay and western Lake Erie) exhibited thymic atrophy (approximately 25% smaller) compared the reference site in the North Channel of Lake Huron. The mass of the bursa of Fabricius and the number of developing lymphocytes in the thymus and bursa were not reduced at contaminated sites. Thymic atrophy was observed in gull chicks from some contaminated sites. The phytohemagglutinin (PHA) skin test, an integrative *in vivo* assay for T cell-mediated immunity, was suppressed approximately 60% compared in gulls from Saginaw Bay, western Lake Erie, and eastern Lake Ontario to the reference colony. The PHA response in terns from Saginaw Bay was suppressed 67% compared to the reference colony in the North Channel of Lake Huron. These biomonitoring data support our previous studies and indicate organochlorine-associated suppression of T cell-mediated immunity in two species of colonial waterbirds at several contaminated sites in the Great Lakes.

PWA238 Optimization, validation and application of biomarkers for the risk assessment of diffusive air pollution. Hamers T.*, Koeman J.H. and Murk A.J. Wageningen Agricultural University, Wageningen, NL. Risk assessment of environmental contaminants is often based on the evaluation of individual compounds. For most compounds present in the complex mixture of air pollution, insufficient data are known on their identity, concentration and toxicity to make a proper risk assessment. Furthermore, such an individual assessment does not take into account the possible combination effects of the compounds. In the present study, bio-assays are optimized, validated and applied to quantify the toxic potency of samples of air and deposition. Both specific bio-assays, to indicate groups of compounds having similar mechanisms of toxicity, and aspecific bio-assays for determination of the overall, general toxicity on groups of organisms such as plants and bacteria are used. The toxic potency of traffic emissions is determined by specific bio-assays for mutagenicity (UMU), Ah-receptor agonism (DR-CALUX), (pseudo)-estrogenicity (ER-CALUX) and by the down-scaled *Vibrio fisheri*-assay for general toxicity. The toxic potency of agricultural pesticide emissions is determined by a microtiter esterase-inhibition assay and by the microtiter *Vibrio fisheri*-assay and daphnid assays. Air filters are sampled near highways and pesticides are sampled in rain water from areas with intense horticulture. All assays are also performed on samples from a nature resort. After adaptation, all bio-assays are apt for quantifying the toxic potency of air pollutants. The results indicate that air pollution with mutagenes, Ah-receptor agonists and IC-inhibitors along the highway is not much higher (< 3-fold) than in a background area. During periods with relatively polluted air (Eastern Wind) this difference was even less. Results of the esterase-inhibition assay for insecticides corresponded very well with chemical analyses on organophosphorus and carbamate compounds in the same samples.

PWA238A Stress response in the Oyster (*Crassostrea virginica*) Exposed to PAH-Contaminated Sediments. Cruz-Rodríguez, L.*, F-L., Chu Virginia Institute of Marine Science, School of Marine Science, Gloucester Point, VA 23062. PAH-contaminated sediments (PAH-CS) were used to determine whether environmental contaminants will induce stress protein response (hsp70) in the eastern oyster *Crassostrea virginica*. Heat shock proteins are highly conserved proteins involved in normal cellular activities but also known to be produced when the cells are challenged with certain environmental stimuli such as high temperature, toxic chemical exposure, and xenobiotics making them a useful marker of exposure to those challenges. Western Blot and Slot Blot format were used to assess the hsp70 expression in the oyster. Exposure of oysters (*C. virginica*) to a shock temperature (33°C) for 1hr and then returned to their original maintenance temperature (13°C) for 2 days, showed that there was an induction of heat shock proteins (hsps). To determine whether environmental contaminants will produce a similar response, oysters were exposed to PAH-CS from Scuffletown Creek, located on the southern branch of the highly contaminated Elizabeth River, Virginia, U.S.A. Oysters were exposed to 0, 1.0, 1.5, or 2.0 g of PAH-CS and the stress response was analyzed after 5, 10, 20, and 40 days. Total PAHs concentrations were 628Fg / g sediment. The results of the analysis of hsp70 in gill tissues indicate that exposure of oysters to 1.0g PAH-CS induced the expression of hsp70 significantly through time (ANOVA $p=0.002$). Results from all PAH-CS exposed groups together suggest a time dependency with an overall increase in hsp70 (ANOVA $p=0.03$). The control groups showed no significant differences through time.

PWA239 Utilizing Graphical, Exploratory and Confirmatory Statistical Techniques to Evaluate Effluent Toxicity. Bliss, B.J.*, Markiewicz, A.J., Luxon, M.G., Landis, W.G., Institute for Environmental Toxicology and Chemistry, Huxley College, Western Washington University, Bellingham, Washington. M. C. Harrass, Amoco Corporation, 28100 Torch Parkway, Warrenville, IL. Data collected by wastewater dischargers typically include information about effluent chemical composition and whole-effluent toxicity (WET) tests. We reviewed an extensive data set from an industrial facility that included such information as well as data obtained during Toxicity Identification Evaluation (TIE) activities to identify specific causes of unacceptable toxicity. Our objectives were to determine what statistical techniques would best improve our ability to analyze and interpret such a large data set and if these techniques could enhance our ability to detect toxicants or conditions contributing to effluent toxicity. Graphical tools and exploratory data analysis methods (correlations, clustering, principal component analyses) were applied to both effluent data and data from internal operations. The types of data included chemical composition, flow rates, and toxicity test results from samples taken from production waste streams, wastewater treatment units, and the final effluent. Data evaluations were complicated by the irregular schedule of sampling and analyses, diverse report formats, the variety of data collected, the sheer volume of data, and the intermittent nature of the observed WET toxicity. Further, few types of data were obtained at every occasion; reflecting the investigative nature of the TIE process, but resulting in an irregular data matrix. Chemical composition data were converted to Toxicity Units (TUs) using water quality criteria or published benchmarks. Preliminary analysis suggested interactions between metals and particulate matter, particularly under certain conditions of low flow and elevated hardness. The benefits of using intensive statistical techniques, relative to conventional approaches for evaluating effluent data sets will be illustrated.

PWA240 pH Dependent Toxicity of Five Metals to Three Marine Organisms. Ho K. T*., Kuhn A., Pelletier M. C., U.S. EPA, Narragansett, RI; Hendricks T.L., Portsmouth, VA. Helmstetter, A. SWCA, Phoenix, AZ. The pH of natural marine systems is relatively stable, which may explain why metal toxicity changes with pH have not been well documented. However in toxicity testing ,changes in metal toxicity with pH in marine waters are of concern. During porewater toxicity testing pH can change 1-2 units as porewater is transferred from *in-situ* to a test container. Also, deliberately altering the sample pH is an important Toxicity Identification and Evaluations (TIE) manipulation designed to detect changes in ammonia toxicity. If altering pH also changes metal toxicity, this may confound interpretation of this manipulation. This study demonstrates that pH alteration changes the toxicity of Cu, Cd, Ni, Pb and Zn to *Mysidopsis bahia* (mysid), *Ampelisca abdita* (amphipod) and *Vibrio fischerii* (Microtox solid phase test (MSP)). Changes in toxicity with respect to pH were metal and organism specific. For the MSP assay, as pH decreased toxicity decreased for Pb, Ni, Cd and Zn and toxicity increased for Cu. For mysids, as pH decreased toxicity decreased for Pb and increased for Cu and Ni. For amphipods, Cu demonstrated decreased toxicity with decreasing pH; the toxicity of all other metals to amphipods remained constant. Results of this study indicate that changes in metal toxicity with respect to pH must be considered for porewater testing and TIE interpretation.

PWA241 Determination of Wastewater Treatment Polymers as Toxicants in an Industrial-Chemical Plant Effluent. MacGregor III, R.* and Clayton, L.A., PBS&J Environmental Toxicology Laboratories, Houston, TX. Effluent from an industrial-chemical plant has exhibited repeated and significant short-term chronic lethality to *Pimephales promelas* but not to *Ceriodaphnia dubia*. A toxicity reduction evaluation program focused on identifying effluent ammonia tolerance limits for *P. promelas*. Initial studies observed an average ammonia NOEC of 6.25 mg/L in effluent as compared to 19.5 mg/L in synthetic hardwater. These data indicated that there were characteristics of the effluent that enhanced the toxicity of ammonia to *P. promelas*. Phase 1 toxicity characterizations were performed on two separate effluent samples that had exhibited significant lethality after ammonia was reduced to 4 mg/L or less. Both Phase 1 studies indicated that the toxicant was filterable, pH-sensitive and effected by EDTA. These findings were consistant with characteristics of two wastewater treatment polymers in use at the plant. A lack of analytical methods to measure these polymers in the wastewater forced alternative confirmation procedures. Toxicity tests with supernatant from the aeration basin mixed liquor, collected prior to exposure to polymers has shown no toxicity to *P. promelas* whereas plant effluent collected during the same time period resulted in significant lethality to *P. promelas*. Alternate treatment polymers are being examined as well as evaluation of ammonia tolerance in the absence of polymer use.

PWA242 Toxicity Identification Evaluation Tools for Metals in Solid Phase Sediment Toxicity Tests. Norberg-King, T.J.*, Maki, R., and Mount, D.R. USEPA, Duluth, MN. With the increasing use of sediment toxicity tests to evaluate dredged material and other sediments, there is a parallel need to identify the causes of sediment toxicity. This need is particularly critical because of its implications for management of toxic sediments; management alternatives for sediments with ammonia toxicity (a common sediment toxicant) are clearly different from bioaccumulative organic contaminants. Toxicity Identification Evaluations (TIE) methods have proven valuable for characterizing and identifying toxicity in effluents and receiving waters. Most sediment TIEs use porewaters, but testing extracted or isolated porewaters may overestimate or underestimate the toxicity of certain compounds. Many sediments contain metal contaminants. Experiments to remove or detoxify heavy metals in sediments have been performed using cation exchange resins, zero-valent metal catalyzation, and iron and sulfide salt additions to produce AVS. Experiments to perform a toxicity characterization of sediment toxicity with Cd, Zn, and Ni spiked sediments and field-collected metal contaminated sediments were conducted using zero-valent metal catalyzation, and iron and sulfide salt additions to produce acid volatile sulfide (AVS), Ambersorb resin and zeolite resins. Subsequent toxicity tests were conducted using the freshwater organisms in 10-d solid phase tests to determine the efficacy for metal removal. The survival of *Hyalella azteca* and the growth and survival of *Chironomus tentans* was assessed. Utility, limitations and interferences of the characterization TIE steps and methods will be demonstrated.

PWA243 Novel Steps in Identifying TDS Toxicity in a POTW Chronic Toxicity Identification Evaluation. Dawson, T.D., Integrated Laboratory Systems, Duluth, MN, Norberg-King, T.J.*, USEPA, Duluth, MN, Lott, K.G., and Deadrick, M., Integrated Laboratory Systems, Duluth, MN. Chronic toxicity in a wastewater treatment plant in Georgia was indicated based on whole effluent toxicity tests with the invertebrate, *Ceriodaphnia dubia*. A toxicity identification evaluation (TIE) was initiated. Phase I TIE characterization tests were not definitive in significantly reducing whole effluent toxicity and subsequent tests indicated total dissolved solids (TDS) were contributing to the toxicity. Mock effluents (ionic composition similar to the effluent) were tested to confirm the TDS levels found in the whole effluent were toxic to *C. dubia*. Chemical analyses and laboratory toxicity data for NaCl identified chloride as the ion responsible for the TDS toxicity. Phase II and Phase III TIE work with TDS have shown that it is difficultto isolate cations or anions from the effluent matrix without producingartificial toxicity. Novel steps in this TIE were the addition of cations (calcium)as an alternative TIE method. These calcium additions reduced toxicity of the effluent and mock effluents. Additions of ~90 mg/L chloride increased toxicity in both the mock effluent and the original whole effluent and these tests confirmed the role of chloride as the toxicant. Species sensitivity comparisons using *Daphnia magna* showed that it was less affected than *C. dubia* by both the whole effluent and the mock effluent which provided more evidence of chloride toxicity. The TIE identified chloride as a major contributor to the effluent toxicity. The TIE data and results of the various TIE steps will be discussed. The methodology used in this TIE illustrate the value of sometimes developing matrix (effluent) specific TIE methods when working with difficult to isolate toxicants.

PWA244 WET Testing Involving Test Waters With High Total Dissolved Solids. Joseph S. Volosin*, Nathaniel Merrill, Kevin V. Brix, and Rick D. Cardwell. Parametrix, Inc., Kirkland, WA. A complication encountered during WET testing for NPDES permits that has gained recognition recently is elevated total dissolved solids (TDS). WET testing for permitees with high TDS effluent or who discharge into receiving waters which are high in TDS can frequently encounter problems with test organism survival which is not related to discharged toxins but rather ion concentrations in the tested water. Similarly, test organisms may be sensitive to specific ion ratios in test waters which may be out of balance even if they are not elevated. When ion related toxicity is encountered during WET testing, no established protocols are currently available to address the problem. Over the past three years, we have conducted a series of studies on high TDS effluents at several sites in the western U.S. These studies have focused on determining whether TDS is the sole cause of toxicity in an effluent, determining the importance of elevated TDS versus ion imbalance for *Ceriodaphnia dubia* and *Selenastrum capricornutum*, and evaluating *Hyalella azteca* and *Ceriodaphnia dubia* acclimated to high TDS waters as alternative test species for high TDS effluents. TDS was toxic to the above organisms in our studies, when cultured under standard conditions. However, *Hyalella azteca* and *Ceriodaphnia dubia* can be acclimated to high TDS. Therefore, alternative or acclimated test species are recommended when evaluating high TDS effluents.

PWA245 Monitoring of reduction toxicity in a Chemical Industry plant production effluent using bioassays with microcrustacean (Daphnia magna). J.L Alberdi*, M.E Sáenz, W.D. Di Marzio and M.C. Tortorelli. Ecotoxicology Research Program, Department of Basic Sciences, National University of Luján. (Bs. As.) Argentina. During eighteen-month reduction toxicity in a Chemical Industry plant production effluent was monitoring. To assess the effectiveness of this process, acute toxicity bioassays with Daphnia magna (Cladocera -Crustacea) were used (USEPA ,1982. Environmental Effects Test Guidelines). The immobility was registrated at 48 hr. The endpoint estimated was EC50-48h using PROBIT or SPEARMAN-KARBER method. The plant production effluent (P.P.E.) contained an active ingredient (A.I.) of parasiticide and a solvent manufactured by the chemical industry. In the first instance were evaluated only the toxicities of A.I. and solvent. Data obtained in the bioassays indicated that of A.I. toxicity was higher than solvent. In a second instance the toxicity of P.P.E. was evaluated. Results indicated P.P.E. EC50-48h were very similar that those obtained in the assays with A.I. In third instance was determinate the toxicity of an effluent develop in a pilot scale reactor by the industry, which A.I. was eliminated by special treatment. The EC50-48h was impossible to determinate, because all the organisms exposed to a 100% concentration of effluent presented mobility. So, the effort was focused to the reduction of concentration the A.I. in P.P.E. Successive samples of plant production effluent were evaluated; these samples were treated in reduction of A.I. concentration, and a significantive decrease in the toxicity was observed in some assays with D. magna. Monitoring results indicated that treatment in reduction toxicity was successful. These assays show the utility of unispecific bioassays with aquatic organisms in reduction toxicity programs of effluents. This was one of the few experience developed in Argentina in this application area.

PWA246 Evaluation of "Green Sand" as a Treatment Option to Remove Manganese from an Effluent. Goodfellow, W.*, Sohn, V., Alexant, J. EA Engineering, Science and Technology, Inc., Sparks, Maryland. Effluent from a groundwater remediation system at a bulk oil storage and distribution terminal has been chronically toxic to *Ceriodaphnia dubia*. Manganese, a naturally occurring substance in the groundwater in that region, has been identified through the performance of a Toxicity Identification Evaluation (TIE) as the principal toxicant in the effluent. Additional characteristics of this effluent are low total suspended solids, total dissolved solids and hardness concentrations. A bench top treatability study was performed to evaluate the effectiveness of "green sand" as a treatment option to remove total and dissolved manganese from the effluent. The "green sand" treatment process also removes beneficial cations such as calcium and magnesium, thus further increasing the toxicity of the effluent associated with low ionic strength. Additional treatment of the effluent was investigated to increase the hardness of the green sand-treated effluent. This paper will present the results of the treatability study, with a discussion of recommendations for addressing the issue of low ionic strength.

PWA247 Assessment of Potential Inhibition to a Wastewater Treatment Plant as part of a Pre-treatment Evaluation. Goodfellow, Jr., W.*, Botts, J.. EA Engineering, Science and Technology, Inc., Sparks, Maryland. As part of a pre-treatment investigation for a large Wastewater Treatment Plant (WWTP), the potential impacts of a significant industrial user (SIU) was evaluated. The wastewater was assessed for the overall impacts to the operations at the WWTP. The impacts of the wastewater to reported odor problem observed at the WWTP, treatment efficiency, and WWTP permit accedences were assessed. In addition, the wastewater was observed to be potentially inhibitory to the BOD measurement used as part of the BOD surcharge used as the basis for fee collection as part of the pre-treatment program. This paper presents the strategy implemented for the pretreatment evaluation. The results of the various assessments including the impact from this wastewater and its relationship to the observed problems at the WWTP and inhibitory effects on the treatment efficiency of the WWTP are discussed. Recommendations as to the use of pre-treatment assessments to evaluating observed WWTP difficulties are also presented.

PWA248 Toxicity Assessment and Treatment Options for Wastewater Associated with Firefighting Training Activities. Novak, L.J.*, Holtze, K.E., ESG International, Guelph, ON, Canada; Young, I.J., Department of National Defence, Hull, QC, Canada; Liu, T., Frazer, J.L., Water Technology International, Burlington, ON, Canada. An evaluation of the toxicity and chemical parameters of wastewater associated with firefighting training at a Canadian Forces Base was undertaken as part of investigations of environmental impacts of chemical waste releases from Department of National Defence operations. The main objectives of this study were to: (i) examine the acute lethal and sublethal toxicity of aqueous film forming foams (AFFFs) and wastewater associated with firefighting activities, (ii) assess if toxicity correlated with wastewater constituents, (iii) evaluate two rapid screening tests (acute and chronic Microtox) to predict responses of standard tests and (iv) assess the effectiveness of reverse osmosis treatment to eliminate toxicity. Rainbow trout (*Oncorhynchus mykiss*) and *Daphnia magna* bioassays were the standard acute lethality tests. The survival/growth test with fathead minnows, survival/reproduction test with *Ceriodaphnia dubia* and growth inhibition test with alga (*Selenastrum capricornutum*) were the sublethal tests. Firefighting wastewater was acutely lethal to both trout and daphnids, but degree of toxicity varied with sampling location. Samples collected from the bottom of containment lagoons were more toxic than those from the surface. The acute Microtox test was the most sensitive acute lethality test, while trout and *D. magna* were the least sensitive. *C. dubia* and alga tests were the most sensitive sublethal bioassays. A comparison of feed and permeate samples indicated that the R.O. treatment system was effective at eliminating toxicity of wastewater collected from the middle of the containment lagoon. Results suggested that the acute Microtox test (IC25 endpoint) could be used effectively to screen the alga toxicity of wastewater associated with firefighting training activities.

PWA249 A Sediment/TIE Approach for Development of Clean-up Goals at an Estuarine CERCLA Site. Tracey, G.A.*, Scott, K.J., Mueller, C., SAIC, Narragansett RI; Svirsky, S., U.S. EPA, Boston, MA. Sediment toxicity and Toxicity Identification Evaluations (TIEs) were conducted on samples collected from an estuarine CERCLA site with high PCBs and metals (As, Cu, Pb, Zn) in order to derive Preliminary Remediation Goals (PRGs) for site remediation. This presentation focuses primarily on the development of PRGs for protection of aquatic life; additional assessments of potential impacts of bioaccumulative CoCs (i.e., dioxins, PCBs) on avian predators or human health are not addressed herein. Nineteen stations throughout the study area and one reference location were selected based on prior bulk sediment data and equilibrium partitioning predictions to obtain sediments of expected toxicity. Fifteen of 20 sediments identified as toxic (*Ampelisca* bulk sediment test, < 85% survival) were selected for subsequent TIE fractionation of porewater to determine CoC classes and associated threshold concentrations for adverse effects. Chemical and toxicity analyses were conducted on C18-(organics removal), EDTA- (metals removal), aeration- (AVS removal) and *Ulva*- (ammonia removal) treated fractions. Results indicate that the toxicity of porewater in surface sediments (0-15cm) to Ampelisca survival and Mulinia larval development are most likely due to copper and not PCBs despite porewater concentrations exceeding 1 mg/L. Under existing conditions, the NOEC was similar among test species and estimated to be 28.7 to 55.4 µg/L. Sediment-based correlations suggest that dioxins in sediment may also induce toxicity when above ~ 100 ng/g TEQ. Finally, high AVS and excellent correlations between SEM and bulk sediment raise concern for enhanced toxicity of Pb, Ni and Zn from oxidized sediments in the surficial sediment microlayer or during resuspension. The lack of comparable correlations for copper are presently not well understood.

PWA250 A Hierarchical Approach to Assessing and Minimizing Environmental Impacts of New Pharmaceutical Manufacturing Processes. Ziegenfuss, P.S.*, Ziegenfuss, M.C., Hannah, R.E., SmithKline Beecham Pharmaceuticals, King of Prussia, PA. A methodology to assess and minimize the environmental impacts and costs of new pharmaceutical manufacturing processes is presented. Production of synthetic organic pharmaceuticals generally involves a series of batch reactions, with one or more waste streams produced for each stage of the process. Initially, process mass balances and waste generation indices are produced for all process streams based on kg of waste per kg of final product. The compositions of all waste streams in the process are initially estimated during early lab scale development and then later refined with analytical data collected during process development. During route development and technology transfer, environmental process assessment of the designated manufacturing route generally involves identification of waste minimization opportunities, followed by solvent recovery and recycling, and biotreatability. As part of the biotreatability assessment, microbial and aquatic toxicity of aqueous wastes are determined. If necessary, pre-treatment processes are developed to reduce the toxicity of aqueous wastes prior to biological treatment. In the case study presented, there were ten organic, three aqueous, and one solid waste (catalyst) streams. Solvent recovery processes were identified for four of the ten organic wastes with potential cost savings and reduction in wasteloads dependent on the manufacturing facilities ability to implement these processes both in the near and far term. Aqueous waste streams were extensively characterized using a suite of chemical parameters, evaluated for toxicity, and assessed for biotreatability. Two of three aqueous wastes were identified to be non-toxic and biotreatable at the predicted production volumes, realizing a potential cost savings of 64 percent compared to on-site incineration. The environmental process development efforts described above resulted in a 62% overall reduction in waste generation for the entire process. An additional 85% reduction in organic wastes will be realized when solvent recovery is implemented in the future.

PWA251 Comparative Toxicity of Produced Water Samples to Three Species and Characterization of Toxicants to the Fertilization Test with *Lytechinus variegatus*. Badaró-Pedroso, C*, EESC, USP, Brazil; Santos, MCF, Depto Fisiologia, IBUSP and EESC, USP, Brazil; Carr, RS, USGS, MERS, TAMU-CC, Corpus Christi,Texas, USA; Rodrigues, P.F.and Sato, MIZ, CETESB, Brazil. Marine toxicity tests with sea urchin *Lytechinus variegatus*, mysid *Mysidopsis juniae* and *Photobacterium phosphoreum* (MICROTOX)were conducted with untreated and chemically treated produced water from Brazil. The treatments consisted of chemical oxidation of sulfides and phenols by hydrogen peroxide and the air stripping technique to reduce oils. The mysid test was the most sensitive with LC_{50}; 96h ranged from 0.92 to 2.66% produced water(PW), followed by MICROTOX EC_{50};15min. (0.55 to 17.2 % PW) and sea urchin EC_{50};80min.(1.82 to 24.29% PW). A phase I Toxicity Identification Evaluation (TIE) study was conducted with the same samples using the *L. variegatus* fertilization assay. The results showed that the most efficient procedures in reducing toxicity were reverse phase chromatography, filtration, *Ulva* spp. addition and chelation with EDTA. Four fraction types were characterized as potential toxicants in the three PW samples: non-polar organics, particulate mater, ammonia and metals. The total oil and greases ranged from 42 to 264mg/L, BTEX from <0.006 to 9.7 mg/L,total solids from 36,690 to 39,280 mg/L, ammonia from 55 to 110 mg/L and metals (Cu, Pb, Zn) from <0.01 to 2.1 mg/L. Research developed with support of CAPES, PETROBRAS and CEBIMar-USP.

PWA252 The Aquatic Toxicity of Evaporator Condensate Produced from the VX Super Critical Water Oxidation Process. Haley, M. V.*, Kurnas, C. W., U. S. Army, Edgewood Research, Development and Engineering Center, SCBRD-RTL, Aberdeen Proving Ground, MD, Turley, S. D. and Burton, D. T., Wye Research and Education Center, University of Maryland, P. O. Box 169, Queenstown, Maryland. The U. S. Army at Aberdeen Proving Ground has investigated an alternative method for disposing of VX (O-ethyl-S(2-isopropylaminoethyl) methyl phosphonothiolate), a chemical warfare agent under the chemical stockpile disposal program. VX was hydrolyzed with sodium hydroxide, placed into a vessel where it was super heated to combustion. The solution was then evaporated to separate the salt from the water. The water produced from the evaporator (evaporator condensate) was subjected to Whole Effluent Toxicity (WET) testing to determine the effects on Selenastrum capricornutum (green algae), *Ceriodaphnia dubia*, (cladoceran) and Pimephales promelas (fathead minnow). Studies were conducted with salt additions to the evaporator condensate and also to distilled water for comparison. It was found that the toxicity associated with the evaporator effluent was due to the lack of dissolved solids. The salts needed for osmotic and ionic regulation were not present in sufficient quantities to maintain normal physiological function.

PWA253 Toxicity in a Blackwater Stream Caused by Naturally Occurring Iron. Specht, W.L.*, Westinghouse Savannah River Company, Aiken, SC; Kelley, R.W., ETT Environmental Inc., Greenville, SC. Blackwater streams, which are common throughout the coastal plain of southeastern U.S., are generally characterized by low pH, low total hardness, and high concentrations of humic and fulvic acids. Toxicity investigations were conducted on water collected from the unimpacted headwaters of Fourmile Branch, a blackwater stream located at the Savannah River Site, a U.S. Department of Energy facility located near Aiken, South Carolina. Water from Fourmile Branch was always chronically toxic and often acutely toxic *to Ceriodaphnia dubia*. In order to determine the cause of the toxicity, a Toxicity Identification Evaluation (TIE) was performed following U.S. EPA methods. The results of the TIE indicate that the toxicity was due to naturally occurring iron, which was present in the water at concentrations as high as 6.2 mg/l.

PWA254 Toxicity Identification Evaluation of Effluent from a Canadian Metal Refinery. Morris, J.R., Laurentian University, Sudbury, ON, Canada; Novak, L.J.*, Holtze, K.E., Wren, C.D., ESG International, Guelph, ON, Canada; Hunt, C.E., INCO Limited, Copper Cliff, ON, Canada. The objective of this study was to identify the substance(s) in INCO's Port Colborne Refinery effluent responsible for causing toxicity to *Daphnia magna* and rainbow trout. The acute toxicity occurrences periodically resulted in non compliance with effluent monitoring regulations. Toxicity Identification Evaluation (TIE) testing conducted in 1996/97 revealed the suspected toxicant(s) were not among those normally associated with effluent from a Canadian refinery. It was concluded that mortality was highly correlated with TDS, however, testing failed to identify which ions best explained toxicity. Several Phase I treatments suggested that unidentified substance(s) also contributed to toxicity. Based on these unknowns and changes in the characteristics of refinery effluent, a second TIE study was undertaken in 1998. It was hypothesized that the effluent may have been toxic because its Ca and K concentrations, unlike Na, were not highly elevated; that is, toxicity may have reflected major imbalances in the Na/Ca or Na/K ratios. Results indicated that zeolite was the only treatment which eliminated toxicity. EDTA, ion exchange columns, XAD and filtration (0.45 μ) had no effect on toxicity. Concentrations of dissolved copper (0.06 mg/L) and cobalt (0.08 mg/L) were reduced by half in post-zeolite treated samples. Nickel (0.25 mg/L) and sodium (2000 mg/L) were only slightly reduced after treatment. Calcium (24 mg/L) and potassium (4 mg/L) increased to 150 and 30 mg/L, respectively. Further testing revealed that addition of Ca alone ($CaSO_4$ or $CaCl_2$) or in combination with KCl delayed and reduced daphnid mortality. It was suspected that the post-zeolite samples may have been non-lethal because the effluent Na/Ca and Na/K ratios had decreased dramatically.

PHA001 Assessing Contaminant Sensitivity of Endangered and Threatened Aquatic Species. Dwyer, F.J.*, Hardesty, D.K., Henke, C.E., Ingersoll, C.G., Sappington, L.C., Whites, D.W. Environmental and Contaminants Research Center, USGS, Columbia, MO. Federal agencies need to assure that chemical use regulations comply with the Endangered Species Act and that these regulations are protective of threatened and endangered (listed) aquatic species. The wide use of pesticides and other commercial chemicals potentially poses a risk to listed species since, by definition, the distribution of listed species is limited and further adverse effects on these populations could lead to extinction. Listed species may not be protected, or unnecessary regulatory programs may be implemented, if the sensitivity of listed species is not evaluated. By evaluating the sensitivity for a number of listed species, it may be possible to make generalizations regarding the protection provided through standard regulatory programs. This research project had two objectives: (1) determine the relative sensitivity to contaminants of listed species using standard acute toxicity tests; and (2) determine the degree of protection afforded listed fish species through the use of standard effluent test procedures. We conducted static acute toxicity tests with two surrogate species, rainbow trout and fathead minnows, and 12 listed species using five chemicals - carbaryl, copper, 4-nonylphenol, pentachlorophenol and permethrin. Toxicity tests were also conducted using standard effluent test procedures with *C. dubia*, fathead minnows and five listed fish. Effluent tests were conducted with ammonia and a chemical mix. Preliminary results from the first study indicate, that in general, LC50s for listed species were more similar to the LC50s for rainbow trout than fathead minnows. For the effluent procedures study, the fathead minnow seven day growth and survival test appears to be a reliable estimator of effects to the listed species tested to date.

PHA002 Comparative Sensitivity of Three Species of Unionid Mussels to a Suite of Toxicants: Glochidia and Juvenile. Keller, A.E.,* and Millar, K., USEPA, Athens, GA; Dorn, L., Mantech Environmental, Athens, GA. Data used to establish water quality criteria in the 1980s contained many values for fish, zooplankton, insects and algae. However, there were few data for freshwater molluscs, mostly Corbicula. As the decline in native freshwater mussels (Family: Unionidae) continues in the eastern United States, questions about what role aquatic contaminants may be playing remain unanswered. The USEPA has recently focused resources on addressing this issue. A series of toxicity tests were performed on glochidia of three species of mussels: *Medionidus conradicus, Actinonaias pectorosa* and *U. imbecillis* to evaluate their relative sensitivity to PCP, SDS, NH_3, anthracene and Cd. The 48-h LC_{50}s for these species were compared to those of other aquatic organisms. Additionally, 6-10 day old juveniles of these three species were tested to the same chemicals in 96-h static tests. Mussel sensitivity was greater to PCP, NH_3 and Cd than to the other two toxicants. Such comparisons among related species and to more typical test organisms will be useful in evaluating the protectiveness of current water quality standards relative to unionid mussels.

PHA003 Aquatic Toxicity and Efficacy of Potassium Permanganate for Zebra Mussel Control. Kent, D.J., The Weinberg Group, Washington, D.C. Since their accidental introduction in 1986, zebra mussels have been an increasingly difficult burden on potable water intake systems. While initially centered in the Great Lakes regions, zebra mussel infestations have expanded their geographic area and have the potential of eventually infesting up to two-thirds of the United States. Of the many forms of chemical control attempted, one of the more promising is the use of potassium permanganate. Potassium permanganate is an oxidizing agent already safely used in water intake pipes of municipal drinking water facilities and other industrial facilities to remove tastes, odors, iron, manganese, and color. It is rapidly reduced in the treatment process and thus has virtually no residual discharge to the environment. Evidence demonstrating that it is also highly effective in the control of zebra mussels in water intakes led to its recent registration under FIFRA for biocidal uses. Laboratory data and case studies show that potassium permanganate's zebra mussel control efficacy can be accomplished at concentrations which are comparable with those already effective for the traditional long-standing non-biocidal uses. The advantages of potassium permanganate over other control chemicals include the lack of THM formation, rapid reduction so no significant residual releases to the environment, and a long history of safe use in potable water facilities.

PHA004 ***In situ* Exposures of Asiatic Clams (*Corbicula fluminea*) and Mayflies (*Hexagenia limbata*) to Assess the Effects of Point and Nonpoint Source Pollution.** Brooker, J.A. and Burton, G.A., Jr.. Institute for Environmental Quality, Wright State University, Dayton, Ohio. *In situ* and laboratory tests of sediment and surface water were conducted using *Corbicula fluminea* and *Hexagenia limbata*. The effects of organisms exposed for 1 to 8 weeks in streams receiving a variety of point and nonpoint source inputs were compared to organisms at reference sites. Exposures partitioned water and sediment related stressors. The usefulness of artificial streams was also examined in simultaneous exposures. Responses of these organisms were compared to the midge *Chironomus tentans*, the amphipod *Hyalella azteca*, and the minnow *Pimephales promelas*. Results showed each exposure system to possess unique information and advantages useful for deciphering dominant stressors and their sources.

PHA005 Analysis of inorganic and organic contaminants in freshwater mussels from the Big Sunflower River, Mississippi. Tatem,H.E.* USAE Waterways Experiment Station, Vicksburg, MS; Lefkovitz,L.F. Battelle/Duxbury, Duxbury, MA; Simmers,J.W. USAE WES, Vicksburg, MS. Nine species of freshwater mussels from the Big Sunflower River were chemically analyzed for three classes of contaminants, including metals, pesticides and PCBs. Mussels were collected from eight sites, from river miles 34.5 to 150 (near Cleveland, MS). The metals Cd, Hg, Pb, Se, Ni and Cr, were found at concentrations generally < 3.0 ppm dry wt. Hg concentrations were generally < 0.2 ppm dry wt. The animals did not contain PCBs, analyzed as Aroclors, at the 5 ppb detection limit. Many pesticides, however, such as toxaphene, DDT compounds, chlordane and dieldrin were found in the mussel tissues. Most of the concentrations were in the < 0.1 ppm wet wt. range however some of these mussels contained toxaphene at concentrations > 0.35 ppm wet wt. The metals and pesticides found were not specific to any one site or mussel species but were evenly distributed. Statistical analyses of the data were used to determine whether the contaminants exceeded USFDA action limits or USFWS predator protection limits. There was a proposal to harvest these animals for their shells and to use the tissues for animal feed. Based on these analyses it was recommended that the tissues not be used as animal feed.

PHA006 Assimilation of Particulate Associated Metals by *Mercenaria mercenaria:* Influence of Bacterial Exopolymer Particulate Coatings. Varlik, B*. Decho, A.W. University of South Carolina, Columbia, South Carolina. The infaunal suspension feeding clam *Mercenaria mercenaria* ingests a range of particles including phytoplankton, suspended sediment particles as well as bacteria. Much of this particulate material is enveloped within mucous exopolymers (EPS) produced by microorganisms. The binding of certain metals may condense EPS coatings, and provide a potential mechanism for the survival of ingested bacteria passing through the digestive tracts of consumer animals. We investigated this process using the EPS-producing marine bacterium *Alteromonas atlantica* and *M. mercenaria*. Assimilation efficiencies (AE) by *M mercenaria* of metals (^{109}Cd & ^{51}Cr-III) and ^{14}C were determined by pulse-chase feeding experiments using radioisotopes. The digestive processing of food material by bivalves follows a "slow" and "fast" pathway. Assimilation of Cr-III by *M mercenaria* is depended on the food type (i.e., sediment suspension (SS) vs. bacterial cells (B)) when present at relatively low concentrations (i.e., ng/ml). This, in part, relates to differential processing of food through the two digestive pathways. Our results indicate significant differences in the digestive processing of different particles types (i.e., sediment vs. bacteria), and in the assimilation of associated Cr-III by the clams B%AE = 25%> SS %AE = 7%. The binding of higher concentrations (mg/ml) of Cr-III or Cd to exopolymer coatings of bacteria and sediments influences the uptake of metals sorbed to these particle types. These results are important for three reasons: 1) Variability observed in metal assimilation by clams, in part, may be influenced by differential digestive processing of the food. 2) Binding of low concentrations of metals to exopolymers may enhance metal uptake associated with the exopolymer and/or particulate material enclosed within the exopolymer. 3) Binding of higher concentrations of metals to exopolymers may decrease the digestibility of particulates enclosed within, and of the metal uptake associated with those particulates. Metal-laden exopolymers may make bacteria less susceptible to digestion.

PHA007 Lethal effects of cadmium in the american oyster *Crassostrea virginica*. Barrera-Escorcia, G. I. Wong-Chang, X. Guzman-Garcia and P. Ramirez-Romero. Universidad Autonoma Metropolitana Iztapalapa, Mexico. It was selected the american oyster Crassostrea virginica to evaluate damage of expossure to cadmium. The final objetive is to determinate physiological effects. In this phase there were obtained the LC50 in adults. The organisms were collected from Mandinga Lagoon, situate 10 Km al Southwest of the Port of Veracruz. The oysters were acclimated for two weeks in laboratory. The bioassay conditions were: D.O.>4 mg/l, Salinity 22‰, 24*C a,d pH 7.99. The organisms were expossed to 5, 10, 20, 30, 40 and 50 mg/l of cadmium as cadmium chloride. The LC50 was 12.5 mg/l. It was observed that the close of valvae were slow in oyster expossed to lower concentrations. The organisms were closed almost all the time in the higher concentrations. The oyster C. virginica is common in mexican lagoons of the Gulf of Mexico. There are studies of pollutants acumulation, but evaluations about the resistence of the oyster to pollutants are escarce in Mexico. The LC1 was 312 ug/l, and it permits the selection of sublethal concentrations to be used in evaluation of physiological effects. It is proposed 100 and 200 ug/l of cadmium to be probe. The maximum limit permited in Mexico for domestic emissions to estuarine environments is 100 ug/l, but the biological effects have a poor evaluation yet. It is necessary to evaluate effects at sublethal level to discuss the limits with a biological criterion.

PHA008 Apple Snail (*Pomacea paludosa*) Responses to Several Pesticides in Acute Exposures. Call, D.J.*, Lake Superior Research Institute, University of Wisconsin-Superior, Superior, WI, U.S.A.; Vyas, N., Patuxent Wildlife Research Center, U.S. Geological Survey, Laurel, MD, U.S.A. Juvenile apple snails (*Pomacea paludosa*) hatched from eggs collected in Florida were exposed to chemical pesticides from several classes and to a microbial pesticide, *Bacillus thuringiensis* (*Bt*), in acute toxicity tests. Exposures were conducted in flow-through diluter systems with continuous renewal of the pesticides at five duplicated exposure levels. Pesticide concentrations were measured daily. EC50 values (96-hr) for the organophosphate insecticide, diazinon, based upon death plus morbidity (immobilization) were 2,950 μg/L for tests starting with 1-day-old snails; and 3,270 and 3,390 μg/L for two tests starting with 7/8-day-old snails. The 96-hr EC50 for 7/8-day-old snails exposed to the inorganic fungicide and algicide, copper sulfate, was 7.42 μg Cu^{+2}/L. The synthetic pyrethroid insecticide, esfenvalerate, was not acutely toxic to 1-day-old apple snails at concentrations up to 1.88 μg/L, the highest concentration attained in the test system. Due to a rapid loss of the fungicide, folpet, from the test water, concentrations were measurable only with a single-cell, high-flow exposure, during which 100 percent of the test animals were killed or affected in 48 hr at a mean concentration of 251 μg/L. *Bt* was not acutely toxic to 1-day-old snails at concentrations as high as 9.2×10^5 colony-forming units (CFU)/mL or to 7-day-old snails at concentrations as high as 8.4×10^5 CFU/mL. These concentrations of *Bt* exceed recommended field application rates for aquatic insect control.

PHA009 The Effect of Cd on development of a freshwater gastropod. Kraly, J.A.*; Roesijadi, G. University of Maryland, Chesapeake Biological Laboratory, Solomons, MD. The developmental toxicity of Cd was investigated in the freshwater gastropod *Biomphalaria glabrata*. *B.glabrata* is a self-fertilizing hermaphroditic species that can also cross-fertilize. Eggs are laid enclosed in gelatinous capsules, and development is direct; individuals hatch as juvenile snails. Criteria for normal development included 1) the first signs of embryonic movement, larval heartbeat, shell growth, and antennal extension and 2) hatching. Eggs were exposed to Cd concentrations ranging from 0.0 to 0.2 μM. A single experiment designed to examine the onset of developmental abnormalities showed no effect of Cd, with the exception of first indication of antennal extension, which correlated with subsequent hatching success. This and four other experiments also showed significant effects on survival and hatching success. Survival curves showed high and concentration-dependent sensitivity to Cd toxicity, with mortalities prior to the hatching event occurring at 0.1 µM and higher (e.g., 100% survival at 0.05 µM versus 80% at 0.1 µM). With hatching, inhibition of hatching was also first observed at 0.1 µM and higher. However, in this case, the concentration-response relationship was steeper and suggested a threshold effect between 0.05 and 0.1 µM Cd (100% hatching at .05 µM versus 20% at 0.1 µM). The mean time to hatch increased at 0.1 µM Cd and higher, as well. Thus, development *B. glabrata* was sensitive to Cd, with an abrupt increase in sensitivity at the concentration of 0.1 µM Cd. Ongoing experiments are examining effects on reproductive output of snails when first sexually mature after lifetime exposure to Cd in addition to survival and hatching.

PHA010 Ecological half-life of ^{137}Cs in largemouth bass (*Micropterus salmoides*) in a contaminated nuclear reactor cooling reservoir: Pond B of the Savannah River Site. Peters, E.L., Chicago State University, Chicago, IL. Pond B is a former nuclear reactor cooling reservoir located on the U.S. Department of Energy's Savannah River Site (SRS) near Aiken, SC. In 1964, R reactor was shut down and the fuel assemblies removed to a water-filled storage basin located near the discharge canal for the hot water effluents. A faulty fuel element contaminated the water in the basin, and approximately 5.7 TBq of ^{137}Cs was released into the pond. More than 30 years later, largemouth bass (*Micropterus salmoides*) from this lake continue to exhibit high ^{137}Cs body burdens (currently about 1 kBq kg-1 wet mass: three orders of magnitude greater than background levels), making them exceptionally suitable for long-term measurements of ^{137}Cs elimination. Sporadic measurements of ^{137}Cs body burdens of Pond B bass have been conducted from 1964 to 1996, and these measurements allow for exceptionally good estimation of the ecological half-life ($T_{1/2}$) of ^{137}Cs in this system. These data indicate two phases in the ecological half-time. For the first 20 years after reactor shutdown, the $T_{1/2}$ was 57 yr (greater than the physical half-life of ^{137}Cs), indicating an increase in bioavailability. From approximately 1983 until the present, however, the $T_{1/2}$ was only 13 yr, indicating that physical sequestration of the radionuclide (presumably due to sedimentation) was accelerating the removal of ^{137}Cs from the system. If the current trend continues, ^{137}Cs concentrations in the bass will decline to levels acceptable for dietary consumption within 60 years. This illustrates the possible long-term patterns of ^{137}Cs bioavailability in other contaminated systems (e.g., lakes contaminated by the Chornobyl nuclear accident).

PHA011 An Integrated Approach for Understanding Watershed Dynamics and Making Management Decisions: The Bailey Creek Case Study. Sammons, T.I.*, Fluor Daniel GTI, Greenville, SC, Hennigar, H.F. The Information Broker, Inc., Saluda, VA, Marcus, J.M., Fluor Daniel GTI, Greenville, SC. The purpose of this study was to determine the overall environmental quality of Bailey Creek through an assessment of landscape ecology, biological inventory, and the physical/chemical nature of the drainage basin. This study was performed in support of an investigation of several contaminated sites within the watershed.. The landscape ecology assessment consisted of a review of topographic and drainage maps, and aerial photography to ascertain drainage patterns. Land usage was field-verified. A habitat assessment, macro invertebrate survey, and fish community structure surveys were conducted at six biological stations. Fifteen water quality/sediment sampling locations were established for organic and inorganic compounds as well as general water quality parameters. The freshwater clam *Corbicula ssp.* was used as a bioaccumulation organism and was maintained for 30 days. Finfish samples were also collected and analyzed as part of the fish community survey. Bailey Creek drains approximately 2,532 acres of the Ft. Lee complex and can be divided into ten (10) drainage compartments which range from highly developed to forested woodland. The aquatic biological assessment indicated that Bailey Creek is at the borderline of "supportive" and "partially supportive" of healthy biological communities with low macro invertebrate taxa richness and a fish population of tolerant species characteristic of a degraded system. Surface water and sediment analyses indicate that low levels of contamination are present. Overall water quality was observed to be poor. The bioaccumulation study indicated that several organic compounds are being accumulated by filter feeders, albeit at very low levels. The results of the biological assessment indicate that Bailey Creek is under considerable stress from constant pulsing of storm water through the system. Because of this, the biological communities are poor to moderate. There do not appear to be significant chemical stressors in the surface water or sediments related to contaminated sites located in the drainage basin. The principal stressor in the Bailey Creek System appears to be storm water runoff and sediment transport. These results were used in the Fort Lee Master Plan to control sediment input into the Bailey Creek system.

PHA012 Building and Communicating Complex Scientific Research Supporting Regional and Watershed Ecological Risk Assessment. Cormier, S.M.*, Schubauer-Berigan, M.E., USEPA, Cincinnati, OH, S. Gordon, Ohio State University, Columbus, OH, M. Smith and D. White, Ohio EPA, Columbus, OH, S. Dyer, Procter & Gamble, Cincinnati, OH, and S.B. Norton, USEPA, Washington, DC. The completeness and quality of ecological risk assessment requires integration of diverse scientific disciplines especially at regional and watershed scales. Such a watershed-based ecological risk assessment in the Big Darby Creek, Ohio necessitated not only a study of the watershed, but also of other watersheds in the Midwest so that quantitative models could be developed to diagnose stressors and project future risk. This approach also allowed the Big Darby Creek watershed to be evaluated from a regional and statewide perspective. This paper introduces a series of studies undertaken to understand watershed issues in Ohio; it highlights how each element contributed to the entire assessment process. Individual research elements are also presented separately at this conference to provide a greater level of detail. Some topics include the ecological risk assessment process and watershed scale analysis, relationships among land use and in-stream habitat condition, and diagnostic signatures from fish and benthic macroinvertebrate assemblages. The ecological risk assessment provided the focal point for designing independent and dependent collaborative projects that will effectively advance ecological science.

PHA013 Using Biological Data to Evaluate Status and Trends in a Watershed. Schubauer-Berigan, M.E.*, Hopkins, J., Cormier, S.M., USEPA, Cincinnati, OH, and Smith, M., Ohio, EPA, Columbus, OH. Assessment of watershed ecological status and trends is challenging for managers who lack consistently sampled data or monitoring programs developed from a watershed perspective. This study investigated potential analytical approaches for assessing status and trends using data collected by Ohio EPA as part of state requirements for reporting stream quality and managing discharge permits. Three time periods, 1979-81, 1986-89, 1990-93, were analyzed for the mainstem of Big Darby Creek, Ohio. Analysis of variance using Tukey's multiple comparisons procedure showed significant differences among time periods for six fish metrics. In addition, significant positive trends were observed for four metrics plus the IBI score, and negative trends for two metrics. An analyses of a subset of sites paired by location and sampled over the three periods reflected findings using all available data for the mainstem. Patterns among time periods were similar for paired sites and for the complete data set, suggesting the sampling was not biased among the three time periods. For benthic macroinvertebrates, five metrics plus the ICI score had significantly positive trends and one metric, a negative trend. A comparison within the 1988-1992 period consistently showed significantly higher quality scores for Big Darby Creek mainstem than for Hellbranch Run, the easternmost sub-watershed. For many fish metrics, Little Darby was not significantly different from Big Darby, but the overall IBI score variable was higher for Big Darby. The consistency of paired and non-randomized data suggest that either type of data might be judiciously used for watershed assessment of high quality streams. Results indicate that overall biological condition of the mainstem of the Big Darby Creek Watershed has significantly improved since the early eighties.

PHA014 Environmental Education: a Hands-on Approach to Watershed Management. Landy, C.*, P. Scheuerman, L. Curtis, and T. Oppewal, East Tennessee State University, Johnson City, Tennessee. Studying watersheds and the ecosystems they support helps us understand the connections between resources and people. Field laboratories provide the means of learning by inquiry while supplying critical data. Watershed education programs use teachers, students, and agencies to solve water problems and educate target groups about water quality issues. The objective of this project was to develop an environmental education program at the high school level. This program is unique because the curriculum incorporates subjects such as math, earth science, and language arts. It is designed to: (1) help students identify problems in their watershed, (2) combine classroom activities with outdoor applications, (3) provide data from biological and chemical monitoring to be used in baseline studies and restoration projects, (4) involve agency partners to provide technical expertise and funding. Watersheds are used as field laboratories for monitoring water quality and aquatic life. There is direct application of science concepts to the environment, and the field studies contribute useful data about local watersheds. The goal is for students to gain real world experiences, improve problem-solving skills, and have a greater respect for water resources. They also develop mapping skills, learn about land use practices and water quality monitoring. Partnerships are formed between schools and communities to adopt local watershed areas. Working together, they can identify pollution problems and find solutions to help improve water quality. Four teachers have already pilot-tested the program in Northeast Tennessee and Southwest Virginia. Eight teachers in Northeast Tennessee have attended a training workshop and are currently implementing the program. Results of program evaluations and teacher surveys suggest that this type of watershed monitoring program is valuable in environmental education.

PHA015 A Management Strategy for Contaminated Sediments in New Jersey Marine Waters. Douglas, W.S.*; DiLorenzo, J; McDonough, F.M.; New Jersey Maritime Resources, Trenton, NJ. Historical industrial activities in the Port of New York and New Jersey have resulted in moderate to severe chemical contamination of the sediments in New Jersey marine waters. In recent years, increasingly sensitive biological and chemical testing of Port sediments has shown that the chemical contamination is bioavailable in many areas. These findings resulted in the closure of the Mud Dump for dredged materials in 1997. Efforts have been underway since 1992 to find alternatives to ocean disposal of dredged materials as well as finding ways to reduce contaminants in harbor sediments. As part of this effort, NJ Maritime Resources has compared available sediment data for the harbor to current NJ State guidelines for cleanup of contaminated soils and sediment quality criteria for ecological health (ERL/ERM). This information suggests that while harbor sediments in some areas are likely a threat to estuarine ecosystem health, the levels of contamination are such that the material is suitable for upland disposal or beneficial reuse as landfill cover, strip mine remediation, or brownfields reclamation. In addition, comparison of surface sediment quality to possible sources has indicated that reduction or elimination of combined sewer overflows in conjunction with remedial dredging followed by sediment decontamination will likely help reduce the immediate threat of contaminated sediments to the harbor estuary. An overall strategy for mid and long-term management of contaminated sediments in the NY/NJ Harbor Estuary is presented.

PHA016 Utilization of Wetland Core Profiles to Determine the Effect of Land use on Contaminant Input to the Elizabeth River, Virginia. Kimbrough, K. L.*, Dickhut, R. M., Virginia Institute of Marine Science, School of Marine Science, College of William and Mary. The Elizabeth River watershed has been stressed by inputs of polycylic aromatic hydrocarbons (PAHs) for decades. Industrial inputs from factories and shipyards resulted in the river becoming one of the most contaminated in the Chesapeake Bay. Regulations and plant closings have curved the input of PAHs from point sources. Present threats to the aquatic environment come from a suite of diffuse and point sources that are deposited by atmospheric deposition, resuspension and runoff. PAH fingerprints will be used to determine the predominant sources threatening the aquatic environment and to link development in the watershed, urbanization and industrialization, to contaminant input. This should elucidate practices that are detrimental to the watershed and show where efforts of remediation should be focussed. A wetland sediment core was taken from Steamboat creek, which drains a residential/industrial watershed. Core characterization consisted of elemental analysis, ^{137}Cs dating, grain size analysis, loss on ignition and PAH concentration determination. Core profiles reveal the history of PAH input to the river and related changes in the watershed. In addition, PAH characterization shows changes in petrogenic and pyrogenic sources.

PHA017 Constructed Wetlands for Treatment of Non-Point Source Pollution. Harre, B., NFESC, Karrh, J., NFESC, Karr, L., NFESC, Kornuc, J., NFESC, Nelson, B., NFESC, Tyzzer, R., Trotsky*, J., NFESC EPA studies have shown that more than a third of the Nation's waters are too polluted for basic uses such as swimming and fishing. These studies indicate that contaminated stormwater runoff is the primary cause for this pollution. The contaminants that enter stormwater runoff typically originate from sources that are difficult to control. Consequently, water quality goals of many receiving water bodies have not been met. Control and treatment of contaminated stormwater runoff is therefore crucial to the quality of our nation's valuable water resources. DoD facilities are often located near areas of sensitive habitat, such as estuaries, rivers, and lakes, where water quality is being impaired. The chemical and physical properties of the contaminants are highly variable, thus requiring a multi-faceted approach to remove them. An approach to stormwater treatment is diverting the runoff into a wetland. Wetlands inherently posses a multitude of contaminant removal mechanisms necessary for treatment of the variable nature of the contaminants in stormwater. In addition, wetlands are self-sustaining and require little operation and maintenance expenditure. While existing wetlands are considered off-limits for treatment in regulatory terms, new wetlands can be constructed which mimic natural wetlands in their water-purifying properties and high cost-effectiveness. The Naval Facilities Engineering Service Center has constructed a tidal-zone wetland at Naval Amphibious Base Little Creek in Norfolk, Virginia to treat stormwater runoff. We use data from this wetland to optimize the operating conditions and maximize the effectiveness of constructed wetlands at other DoD locations. This poster will detail our research and portray the benefits of constructed wetlands.

PHA018 Methodology for Determining Impaired Waters by Toxics for the 303(d) List. Jiapizian, P.B.*, Kroll R.B., George J.W., and Eskin, R.A. Maryland Department of the Environment, Baltimore MD. A tiered decision pathway is being developed to objectively determine water quality limited segments (WQLS) for the State's 303 (d) List (List of Impaired waters). The tiered approach will compare various types of chemical and biological data to regulatory criteria and available reference screening levels. The first tier in evaluating potential impairments to water bodies will examine exceedances of numeric ambient water quality criteria by water column data. In the absence of ambient water column data or water quality criteria, a second tier weight-of-evidence approach will be used to determine WQLS. Three categories of data will be used for this approach: sediment contamination data, biological community data, and ambient water column and sediment toxicity data. Segments will be listed as WQLS only if potential impairment due to sediment chemical contamination is also confirmed with either ambient toxicity testing or biological community impacts. (This is an abstract of a proposed poster and does not necessarily reflect MD. Dept. of Environment policy.)

PHA019 Changing players in environmental decision-making: from geo-politics to eco-politics Glicken, J.*; Clark, K.S. ecological planning and toxicology, inc., Albuquerque, NM Relevant stakeholder groups, appropriate and effective decision-making bodies, and the ecosystem scales at which the risk assessments are conducted are all intimately coupled. The stakeholder group(s) will help identify appropriate indicators for the risk assessment, helping to determine the ecosystem scale required. The decision-making body may or may not be isomorphic with ecosystem identified as the target of the risk assessment, or with the full universe of stakeholders in the appropriate functioning of that ecosystem. If it is not, there may be serious questions about the authority and jurisdiction of the decision-making body to implement any decisions it makes relative to the target ecosystem. Movement of environmental decision-making has been increasingly to ecoregion-defined organizations (both formal and informal), and away from geopolitically defined entities such as nation-states and municipalities. This research will explore an instance of such movement in New Mexico, specifically in relation to water management in the middle Rio Grande Valley. Theoretical research on issues of 'environmental security' and environmental decision-making will be combined with ethnographic accounts of water-related decision-making in the target area. The research is anticipated to yield insights into areas of social conflict, as jurisdictions defined by ecology, use, and impact conflict with geopolitical boundaries and established social mechanisms of decision making and enforcement.

PHA020 Effects of PCB 126 on Thymocyte Phenotypes in Chicken Embryos. Grasman, K.A. and Fox, L.L., Wright State University, Dayton, OH. This study evaluated the effects of developmental exposure to a planar polychlorinated biphenyl (PCB) on thymic organ weight and thymocyte numbers and phenotypes in chicken embryos. Exposure to planar halogenated aromatic hydrocarbons (HAHs) causes thymic atrophy and decreases thymocyte numbers. T-lymphocytes develop in the thymus. Helper T-cells express the CD4 molecule on their surface and stimulate both cytotoxic T-cells and antibody-secreting lymphocytes (B-cells). Cytotoxic T-cells express the CD8 molecule on their surface and kill virus- infected cells. In marmosets and rats, planar HAHs decrease percentages of the most immature thymocyte population, CD4+CD8+, and increase percentages of CD8+ cells. Both of these subsets express the $\alpha\beta$ T-cell receptor (TCR). A separate T-cell population expresses the $\gamma\delta$ TCR. This experiment compared effects on thymocyte phenotypes, cell numbers, and thymic weight in chicken embryos. Chicken eggs were injected with PCB 126 (sunflower oil carrier) into the air cell before initiation of incubation. Doses ranged from 0.0512 to 0.8 ng/g egg. Control groups included non-injected and carrier-injected eggs. The thymus was removed on day 20 of incubation (one day before hatch) and homogenized. Thymocytes were stained with monoclonal antibodies to surface proteins and analyzed by flow cytometry. Thymic atrophy was observed at 0.128 ng/g with maximal suppression at 0.8 ng/g. Thymocyte numbers were more variable than thymus mass but were reduced at and above 0.128 ng/g. Preliminary analysis of CD4/CD8 and TCR phenotypes revealed no dose-dependent changes despite decreases in thymus mass and thymocyte numbers at higher doses.

PHA021 The Effects of PCB 126 on Immune Organ Development and Thymocyte Proliferation in Chicken Embryos. Croisant, E.T.*, Fox, L.L., and Grasman, K.A., Wright State University, Dayton, OH. PCB exposure has negative effects upon thymus mass, thymocyte development and T-cell activity. The natural development of thymocytes is critical to their optimal activity as mature T-cells. Lymphoproliferation is a useful tool for assessing immunocompetence. Exposure of mammals to dioxins and PCBs causes suppression of mitogen-induced lymphoproliferation. This study explored the hypothesis that PCB 126 reduces the proliferation of chicken embryo thymocytes. PCB 126 (0.0, 0.08, 0.2, 0.5 ng/g in sunflower oil) was injected into the air cell of chicken eggs at day zero of incubation. Tissues were collected at day 20 of development. Thymocytes were isolated from the thymi, counted and cultured in 96 well plates with or without varying concentrations of the T-cell mitogens phytohemagglutinin (PHA) or concanavalin-A (Con-A). Proliferation was assessed using Alamar Blue, a growth indicator that is reduced to a fluorescent form by the metabolism of proliferating cells. PCB exposure reduced body mass, immune organ mass, organ cell number and increased mortality, deformities and yolk mass. The strongest proliferation in mitogen-treated cultures was at concentrations of PHA above 100mg/500,000 cells. Con-A did not stimulate proliferation. There was an indication of reduced thymocyte proliferation at 0.08 and 0.5 ng/g. In general, no monotonic PCB dose response was observed in thymocyte proliferation induced by T-cell mitogens. A metabolism based indicator dye may not be optimal for measuring the proliferation of thymocytes. Apoptosis occurs in >90% of the thymocyte population *in vivo* and *in vitro*. The dying cells may decrease the reduction of the dye and interfere with assessment of proliferation. Alternate methods should be explored for the measurement of this endpoint.

PHA022 Antioxidant Adaptations in Killifish (*Fundulus heteroclitus*) Populations from PAH-impacted and PCB-impacted Sites. Carey, J. A.*, MacLean, E.D., Di Giulio, R.T., Duke University, Durham, NC. Populations of killifish, *Fundulus heteroclitus*, from Elizabeth River (ER), VA, and New Bedford Harbor (NBH), MA, have been reported to thrive despite high levels of PAHs and PCBs in their environments. Also, fish from these impacted sites have been reported to be more resistant to the acute toxicities of these chemicals than fish from uncontaminated neighboring reference sites. This study provides a preliminary investigation into compensatory mechanisms underlying this apparent adaptation to chemical stress from the perspective of antioxidant adaptations. To test this mechanism, several parameters related to oxidative stress were compared in livers of fish from contaminated versus reference sites. The parameters included concentrations of reduced and oxidized glutathione (GSH and GSSG), menadione-mediated microsomal production of superoxide anion, and activities of glutathione reductase (GR), g-glutamylcysteine synthetase (GCS), ethoxyresorufin O-deethylase (EROD), and cytochrome P450 reductase (P450R). The ER (PAH-impacted) population was observed to have significantly increased ($p < 0.05$) concentrations of glutathione (GSH) and glutathione disulfide (GSSG). After allowing this population to depurate, GSH and GSSG values returned to reference levels. GR activities were significantly greater in ER fish, but no differences were observed in other enzyme activities. In contrast, GSH concentrations were significantly lower in the NBH fish (PCB-impacted) versus reference fish , but no difference was observed in GSSG concentrations. No differences in enzyme activities were observed in fish from NBH. These results provide evidence for a glutathione-based adaptation as a possible mechanism conferring resistance to PAH-impacted populations of killifish. In contrast, no evidence for antioxidant adaptations was observed in fish from a PCB-impacted site.

PHA023 Investigation of the Presence and Function of a Putative Multidrug Transporter in the Grass Shrimp, Palaemonetes pugio. Finley, DB*1, Scott, GI1 and Karnaky, K2. 1National Ocean Service, Charleston, SC. 2Medical University of South Carolina, Charleston, SC. Permeability glycoprotein (P-gp) is a 170 kDa integral plasma membrane transport glycoprotein which is up regulated in some cancer cells exposed to chemotherapeutic drugs. It then imparts protection in the form of multidrug resistance. Most P-gp research has focused on human cancers; however, since many marine organisms thrive and reproduce in polluted areas, recent questions have been posed about P-gp homologous proteins in these organisms and their role in protection against pollution stress. To address this question, an intensive study of the grass shrimp, Palaemonetes pugio, which commonly inhabits South Carolina estuaries, was performed. This study included quantitative monthly monitoring of putative P-gp expression in grass shrimp from different estuaries (urban, agricultural and reference) using the human monoclonal antibody C494. In addition, the intestine of the animal was characterized using electron microscopy and then, using immunocytochemistry, P-gp was localized to the luminal epithelial cells of the intestine and the outer epidermis under the exoskeleton. P-glycoprotein specific transport activity across the intestine and embryonic cell coat was monitored using daunorubicin, a fluorescent substrate of P-gp. Daunorubicin transport was then tested for inhibition with a P-gp specific inhibitor (verapamil) and a common estuarine contaminant (endosulfan). Results suggest evidence for both the presence and function of a P-gp homologous transporter in the grass shrimp, P. pugio.

PHA024 Molecular Characterization of Contaminant-Indicative RAPD Markers: Homology Between Species, DNA Sequence, and Development of Marker-Specific Primers. Theodorakis, C.W.*, Bickham, J.W., Texas A&M University, College Station, TX; Chesser, R.K., Savannah River Ecology Laboratory, Aiken, SC. The objective of this research is to utilize the randomly amplified polymorphic DNA (RAPD) technique to develop population genetic markers that can be used for risk assessment of chronic environmental contamination. Previous research found RAPD markers which were present at a higher frequency in radionuclide-contaminated than in reference mosquitofish (Gambusia affinis) populations from Oak Ridge, TN. These markers will be referred to as "contaminant-indicative markers" (CIMs). In the present study, the same RAPD primers were used to examine populations of G. holbrooki collected from radionuclide-contaminated and reference sites in South Carolina. Three markers were present at a higher frequency in the contaminated populations, and 2 were present at a lower frequency. CIMs from G. affinis were isolated and probed against G. holbrooki RAPD products using Southern blotting. This assay revealed that all the aforementioned G. holbrooki CIMs were homologous to G. affinis CIMs. Four of these markers were then cloned and sequenced. All 4 markers shared certain similarities, e.g. they were all adenosine-thymidine rich (60-75% AT) and possessed the potential for extensive secondary structure. These sequences were used to design polymerase chain reaction (PCR) primers which would amplify only one CIM at a time (typical RAPD-PCR reactions amplify numerous markers simultaneously). Some primers amplified products not only from G. affinis but also from other species (including humans), although more than one PCR product was amplified for each species. Other primers amplified only one Gambusia marker, but did not work with other species. Use of marker-specific primers in population genetic studies could obviate some of the problems (e.g., non-reproducible bands, codominance) inherent with the RAPD technique.

PHA025 Cloning of cDNA and promoter sequences of Cytochrome P4501A from killifish (*Fundulus heteroclitus*). Powell, W.H.*[1], Morrison, H.G.[2], Weil, E.J.[2], Karchner, S.I.[1], Sogin, M.L.[2], Hahn, M.E.[1], Stegeman, J.J[1]. [1] Woods Hole Oceanographic Institution and [2] Marine Biological Laboratory, Woods Hole, MA. Members of the cytochrome P450 gene family 1 (*CYP1*) catalyze the metabolic activation of numerous hydrocarbon carcinogens and various natural compounds. *CYP1* family members have been identified in several vertebrates, including fish, birds, and mammals. Induction of P450 1A gene expression is a hallmark response of many organisms and cells exposed to aryl hydrocarbon xenobiotics. Transcriptional activation of *CYP1A* is mediated through the binding of xenobiotics to the aromatic hydrocarbon receptor (AHR). With its heterodimeric partner ARNT, the ligand-bound AHR binds conserved xenobiotic response elements (XREs) near the promoter of *CYP1A* and other genes. We report here the isolation of cDNA and genomic clones of *CYP1A* from *Fundulus heteroclitus*, a teleost fish common in even highly polluted estuaries on the Atlantic coast. The cDNA encodes a polypeptide sequence of 521 amino acids (predicted molecular weight is 59.3 kDa) which shares 65%-80% identities with known fish *CYP1As*. This sequence includes a putative heme-binding cysteine in a typical P450 signature sequence (FGLGRRRCIG) and an eighteen residue sequence (SDEKIVGIVNDLFGAGFDT) highly conserved among teleost *CYP1As*. The genomic clone encodes an identical amino acid sequence. Consistent with its inducibility by AHR agonists, the *CYP1A* gene contains three consensus XREs (5'CACGCNA3') within 2.5 kb of the putative transcriptional start site. Discrete populations of *F. heteroclitus* vary in their sensitivity to certain xenobiotics, including dioxins, PCBs, and polycyclic aromatic hydrocarbon mixtures. These sequence data provide a valuable tool for the study of genetic variations in *CYP1A* expression and activity in sensitive and resistant populations. These studies may ultimately shed light on the importance of P4501A activity in xenobiotic toxicity.

PHA026 Molecular Characterization of Ah Receptor cDNAs in Birds and Amphibians. Karchner, S.I.*, Hahn, M.E., Woods Hole Oceanographic Institution, Woods Hole, MA; Kennedy, S.W., Trudeau, S., Environment Canada, CWS/NWRC, Hull, Quebec, Canada. The aryl hydrocarbon receptor (AHR) is a ligand-activated transcription factor that mediates the toxic effects of dioxins and other halogenated aromatic hydrocarbons (HAHs) through transcriptional activation of several genes. The role of the AHR in induction of cytochrome P450 1A1 is well characterized, and CYP 1A1 induction has been utilized as a sensitive marker for the activation of AHR-dependent pathways. Studies of CYP1A inducibility in cultured avian hepatocytes have revealed dramatic species differences in the sensitivity of birds to HAHs (Kennedy et al Toxicol. Appl. Pharmacol. 141:214 (1996)). To investigate the basis of observed differences in HAH sensitivity, we have begun to characterize the AHR in several species of birds (White leghorn chicken (*Gallus gallus*), Pekin duck (*Anas platyrhynchos*), Common tern (*Sterna hirundo*) and Herring gull (*Larus argentatus*)), as well as an amphibian (mudpuppy (*Necturus maculosus*)). Partial AHR cDNAs encompassing the helix-loop-helix and PAS domains were cloned and sequenced from these species. Comparison of amino acid sequences in this region indicated a high degree of sequence conservation among these bird species (97% amino acid identity). The percent identity between bird sequences and either mouse or mudpuppy was lower (79%), and the mudpuppy AHR was 74% identical to the mouse AHR. Phylogenetic analysis of these sequences and other AHR cDNAs is an initial step in investigating interspecies differences in the AHR signaling pathway. (Supported in part by Grant ES06272 from NIEHS and by NOAA Sea Grant).

PHA027 Effects of PCB 126 on Primary Immune Organ Development in Chicken Embryos. Grasman, K.A. and Fox, L.L. Wright State University, Dayton, OH. This experiment evaluated the immunotoxic effects of developmental exposure to planar PCBs. Most previous investigations on the immunotoxic effects of PCBs in developing avian embryos have exposed embryos only during the latter half of incubation. To simulate exposure in wild embryos, chicken eggs were injected with PCB 126 (sunflower oil carrier) into the air cell before initiation of incubation. Doses ranged form 0.0512 to 0.8 ng/g egg. Control groups included non-injected and carrier-injected eggs. The thymus and bursa were removed and weighed on day 20 of incubation (one day before hatch). The immune organs were homogenized, and viable lymphoid cells were counted using the trypan blue exclusion method. Probit analysis estimated the LD50 to be 1.04 ng/g. Lymphocyte counts decreased at or below doses causing organ atrophy. Bursa mass began to decrease at the lowest dose of 0.0512. The number of viable cells decreased slightly at 0.0512 and reached a minimum at 0.128 ng/g. Measures of B-cell maturation were reduced at doses lower than those associated with PCB-induced mortality. Thymus mass dropped sharply between 0.128 and 0.32 ng/g, and lymphoid cell numbers in the thymus fell between 0.0512 and 0.128 ng/g. Both organ weight and cell numbers decreased at lower doses in the bursa than in the thymus. This suggests that in chickens the bursa is more sensitive to planar PCBs than the thymus. Doses necessary to reduce the number of viable lymphocytes in the thymus and bursa are at least 1 order of magnitude lower when administered at the beginning of incubation as compared to previously published studies of exposure only during late incubation.

PHA028 Species Specific Recombinant Cell Bioassays: Differences in Sensitivity to Complex Mixtures of Halogenated Aromatic Hydrocarbons (HAHs). Villeneuve, D.L.*, Staffova, K., Kannan, K., and Giesy, J.P., Michigan State University, East Lansing, MI. Fish and mammals have been reported to possess differing sensitivities to certain classes of HAHs. The RLT 2.0 bioassay, which utilizes rainbow trout cells stably transfected with a luciferase reporter gene under control of dioxin responsive elements, was developed in response to the apparent need for fish-specific *in vitro* bioassays. This study tested the hypothesis that the RLT 2.0 bioassay responds differently than an analogous mammalian cell bioassay (H4IIE-luciferase) to environmental samples containing complex mixtures of HAHs. Sediment extracts from a superfund site contaminated with Arochlor 1268 were analyzed using RLT 2.0 and H4IIE-luciferase bioassay. The bioassays predicted different rank orders of potency for the samples tested. In terms of magnitude, RLT 2.0-derived 2,3,7,8-tetrachlorodibenzo-*p*-dioxin equivalents (TCDD-EQ) were up to 10 fold greater for PCB fractions and 100 fold greater for dioxin / dibenzofuran (DDF) fractions (which also contained polychlorinated naphthalenes -PCNs) than corresponding H4IIE-derived TCDD-EQ. H4IIE-derived TCDD-EQ were similar ($\leq$ 2.5 fold difference) to international TEQs (I-TEQ) calculated for DDF fractions based on dioxins and furans alone. When I-TEQs for PCNs were added to dioxin and furan TEQs, RLT 2.0 TCDD-EQs were found to be more similar. Total I-TEQs for the DDF fractions were 3550, 2700, and 3800 pg TEQ/g sediment for samples KS-1, KS-2, and KS-3 respectively. RLT 2.0 TCDD-EQs for DDF fractions were 2730, 19,850, and 2700 pg TCDD-EQ/g sediment, compared to 1160, 250, and 28 pg TCDD-EQ/ g sediment for H4IIE. These results indicate that there are differences in sensitivity between RLT 2.0 and H4IIE-luc bioassay. Specifically, the RLT 2.0 bioassay may be more sensitive to PCNs than the H4IIE-luc assay.

PHA029 Species-Differences in the Inhibition of Fish Serum B Esterases by Organophosphate and Carbamate Pesticides. Huang, T.Y.*, La Point, T.M., Hooper, M.J., The Institute of Environmental and Human Health, Texas Tech University, Lubbock, Texas, Chuiko, G.M., Institute of Inland Water Biology, Russian Academy of Science, Borok, Yaroslava, Russia. Plasma B esterases have been shown to sequester anticholinesterase pesticides in mammals and birds, protecting critical brain cholinesterase from inhibition. Inhibit ance in some species, it may serve an important role for detoxification of organophosphate and carbamate contamination and as a good biomarker. While most fish species contain plasma AChE, its role in detoxification by non-target binding might not be significant due to its low abundance and lower affinity. CaE was found in serum of all tested species except channel catfish. It may be the most important protective enzyme for OPs in those species lacking BChE. The phorothioate oxon, chlorpyrifos-oxon was the mos milar AChE and BChE inhibition cap.

PHA030 Allozyme Analysis in Aquatic Snails From a Mercury-Impacted River. Benton, M.J.*, East Tennessee State University, Johnson City, TN; Trybula, J., Guttman, S.I., Miami University, Oxford, OH. The relationship of environmental mercury exposure on allozyme genotype frequencies in aquatic snails were tested by starch gel and cellulose acetate plate electrophoresis. *Pleurocera canaliculatum* were collected in two consecutive years from two sites upstream and three sites downstream of Saltville, VA, on the North Fork Holston River (NFHR). The NFHR is severely mercury-impacted at Saltville, with mercury levels generally low upstream and decreasing with distance downstream. Thirteen presumptive polymorphic loci were analyzed: EST-1, EST-2, EST-3, EST-4, ALP-1, GPI-1, TPI-1, MDH-1, MPI-1, PEP-2, PEP-3, EUV-4, and ACP-3. In year 1, EST-2, ALP-1, TPI-1, and PEP-3 genotype frequencies differed significantly among sites, while in year 2 only ALP-1 and ACP-3 genotypes differed significantly. When comparing genotype frequencies between sites grouped as upstream and downstream of Saltville, only PEP-3 differed significantly in year 1, while in year 2 MDH-1 and PEP-2 differed significantly. For the two years combined, EST-2, TPI-1, MDH-1, PEP-2, and PEP-3 genotype frequencies differed significantly among sites. Genotype frequencies for TPI-1, MDH-1, PEP-2, and PEP 3 differed significantly between sites upstream and downstream of Saltville. Results suggest that mercury-contaminated conditions may select for some allozyme genotypes, but other influences appear to cause temporal and spatial variation in overall population genetic structure.

PHA031 Single- and Double-Strand DNA Breakage in Aquatic Snails From a Mercury-Impacted River. Benton, M.J.*, East Tennessee State University, Johnson City, TN. The effects of environmental mercury exposure on DNA integrity in aquatic snails were tested using agarose gel electrophoresis. *Pleurocera canaliculatum* were collected from three sites on the North Fork Holston River near Saltville, Virginia, which are known to have relatively low, moderate, and high mercury levels in the sediments. DNA was chemically extracted from the homogenized foot of 16 individuals from each site and subjected to alkaline- and neutral-pH gel electrophoresis. Exposure to alkaline conditions causes the separation of complementary DNA strands. Following electrophoresis, intact single-stranded DNA appears on the gel as a coherent band, while DNA fragments caused by single- and double-strand breakage (i.e., total breakage) appear as a smear below the band. On neutral gels following electrophoresis, double-stranded DNA appears as a coherent band, while DNA fragments due to only double-strand breaks appear as a smear. By densitometrically quantitating relative amounts of DNA in the coherent band and in the smear, a measure of strand breakage can be calculated. By subtracting double-strand breakage from total breakage, an estimate of single-strand breakage can be obtained. Snails from the high mercury contamination site exhibited more single- and double-strand breakage than those from the low and moderate mercury contamination sites. Results suggest that mercury plays a role in environmental contamination-induced genetic damage.

PHA032 Relationship Between Acute Toxicity and Heterozygosity in the Amphipod, *Hyalella azteca*. Eisenhauer, J. B., Brown Sullivan, K., and Lydy, M. J., Wichita State University, Wichita, Kansas. This study examines the relationship between acute toxicity and genetic variability in the amphipod, *Hyalella azteca*. Several studies have shown that characteristics of growth, reproduction, and survival are positively correlated to the amount of heterozygosity within individuals. Only a few studies have examined the relationship between an individual's heterozygosity and tolerance to environmental stressors. *H. azteca* was chosen for this investigation because it expresses genetic variability at a variety of enzyme loci, and it is routinely used in acute toxicity tests. Genotypes for three enzymes, acid phosphatase (ACP) (EC 3. 1. 3. 2), glucose - 6 - phosphate isomerase (GPI) (EC 5. 3. 1. 9), and phosphoglucomutase (PGM) (EC 5. 4. 2. 2), were examined using cellulose acetate gel electrophoresis (CAGE). Acute, 96-hour toxicity tests were performed using zinc as a stressor. The LC_{50} value for zinc was determined to be 435 ppb using the Trimmed Spearman - Karber statistical program. The LC_{70} value (700 ppb), estimated from data plotted on log probit paper, was used for a 96-hour time to death test. Amphipods were examined for mortality at three hour intervals during this test. Differences between genotypes of surviving and non-surviving organisms will be discussed. Also, the relationship between individual heterozygosity and time - to - death will be presented.

PHA033 Immunomodulation and Altered Cytochrome P450 Expression by Clofibrate and Retinoic Acid in *Fundulus heteroclitus*. Baier-Anderson, C.*, Anderson, R.S., Haasch, M.L., University of Maryland Center for Environmental Science, Chesapeake Biological Laboratory, Solomons, MD. There is conflicting evidence on the immunomodulating potential of peroxisome proliferating agents (PPAs). Retinoids are important to immune system function and there is evidence that enzymes induced by exposure to PPAs may alter retinoid homeostasis, which, in turn, could modulate tissue macrophage activity. The purpose of this research is to (1) determine if exposure of fish (*Fundulus heteroclitus*) to all-*trans*-retinoic acid modulates tissue macrophage function; (2) to correlate immunomodulation with lauric acid hydroxylase induction; and (3) to characterize the immunomodulatory potential of clofibrate, a potent model PPA. In this preliminary study, fish were exposed for 24 hours to all-*trans*-retinoic acid (1 mM), clofibrate (0.04 and 0.4 mM) or vehicle. Macrophages from the anterior kidneys were harvested and zymosan-stimulated reactive oxygen species (ROS) production was measured using luminol-augmented chemiluminescence. Liver microsomal fractions were probed for lauric acid hydroxylase induction and cytosolic fractions were probed for nitric oxide synthase (iNOS) induction. The results indicate that all-*trans*-retinoic acid significantly increases, whereas clofibrate decreases, ROS production. Two antibodies were used to probe for lauric acid hydroxylase induction: anti-CYP2M1 and anti-CYP2K1. The antibodies recognized several induced proteins in liver microsomes following exposure to clofibrate and retinoic acid. Similarly, iNOS was induced in liver cytosols, indicating the presence of activated macrophages. These results suggest that environmental exposures to PPAs may cause tissue-dependent alterations in macrophage function. Furthermore, since all-*trans*-retinoic acid activates macrophages, it is possible that altered retinoid homeostasis could be the mechanism by which PPAs exert their immunomodulatory effects.

PHA034 Protection effect of Cd/Zn-Metallothionein in the *in vitro* DNA cleavage induced by copper-1,10-phenanthroline complex. Jianhua Yang, M.S.Yang, R.N.S. Wong. Hong kong Baptist University, Hong Kong, China. Using supercoiled plasmid DNA relaxation assay, the protection effect of Cd/Zn-MT on the DNA cleavage activity of the $[(OP)_2Cu^+]$ complex was investigated. And its possible mechanism of the action for MT was analyzed The DNA cleavage activity of the copper-1,10-phenanthroline complex was negatively correlated to the concentration of MT. The nuclease activity of the complex was gradually inhibited by MT when the concentration of MT was increased from 0.01μM to 1.0μM. At 2.5μM, MT completely inhibited the DNA cleavage caused by the complex. The preliminary results showed that the inhibition may be due to that MT can not only cause the decomposition of the DNA·$(OP)_2Cu^+$] complex but also can block the formation of the DNA·$(OP)_2Cu^+$] complex. In the same experiment, we found cysteine has the same function as MT. MT and cysteine can work synergistically to inhibit the DNA cleavage by the copper-1,10-phenanthroline complex. However, based on the number of the sulphur unit, MT was a 1000-fold more efficient than cysteine in inhibiting the DNA cleavage. These results support that MT might play important role in protection against free radical generating DNA damage.

PHA035 Effect of Individual and Mixtures of Organochlorine Compounds on Human Natural Killer (NK) Cell Function. Loganathan, B.G.*, Owen, D.A., Whalen, M.M., Murray State University, Murray, KY; Warren, T. Murray Calloway Hospital, Murray, Cytotoxicity studies were conducted to determine the effect of selected non-*ortho*-, mono-*ortho*-, di*ortho*chlorine substituted PCB congeners, mixture of the congeners, and Aroclors on human natural killer (NK) lymphocytes function (their ability to kill cancer cells) *in vitro.* Selected chlorinated pesticides including hexachlorobenzene (HCB), 4,4'-DDE and 4,4'-DDT were also examined *in vitro* for their effect on NK cell function. Standard methods were used for human NK cell preparation, chemical preparation, cell viability and measurement of NK cytotoxicity using ^{51}Cr release assay (K562 cancer cell line was used as target cells). The assays were conducted in gelatin media and cell viability was measured beginning and end of the assays. The individual and mixtures treatment concentrations ranged (environmentally relevant concentrations) from 2 nM to 1 μM and exposure period were 1 h, 24 h and 3 days. Additional *in vitro* assays were conducted to determine the impact of selected compounds on the interleukin-2 (IL-2) stimulation of NK cells. The results allowed us to conclude that: (i) PCB-77, PCB-126 and PCB-169 did not suppress NK cell function individually or mixtures even at 1µM level even at 3 days *in vitro* exposure. (ii) PCB-118 and PCB-153 did not alter NK cell function at 400 nM concentration for 3 days. (iii) Three days exposure of HCB + 4,4'-DDE +PCB-126 at 200 nM each did not disrupt NK cell function. (iv) Highly toxic PCB congeners did not interfere with the IL-2 stimulation of NK cell function. (v) Aroclor 1016 and Aroclor 1254 showed significant effect and suppressed NK cell function by 47% and 26 % respectively at 100 ppm exposure concentration for 3 days.

PHA036 Effect of Tributyltin (TBT) on Human Natural Killer (NK) Cell Function. Whalen, M.M., Loganathan, B.G.*, Owen, D.A., Murray State University, Murray, KY; Warren, T. Murray Calloway Hospital, Murray, KY; Kannan, K. Michigan State University, MI. Tributyltin (TBT) is a well known highly toxic environmental pollutant. Immunotoxic effect of TBT on experimental, terrestrial animals are well documented. However, immunotoxic effect of TBT on humans, particularly human natural killer (NK) cell mediated immunotoxic effect is not yet described. NK Cells play a central role in immune defense against viral infections and tumor cells. The objective of this study was to determine the *in vitro* effect of TBT on human NK cell function (its ability to kill cancer cells). Cytotoxicity studies were conducted using 1 h, 4 h, 24 h preincubated human NK cells (of several male and female individuals) with 50 nM, 100 nM and 200 nM TBT to determine the effect on NK cell function *in vitro*. Additional *in vitro* assays were conducted to find out, if interleukin2 (IL2) could protect the cells from the inhibitory effect of TBT. The ability of TBT treated NK cells to bind to tumor cells was also assessed by conjugate formation tests. Standard methods were used for human NK cell preparation, chemical preparation, cell viability and measurement of NK cytotoxicity using ^{51}Cr release assay (K562 cancer cell line was used as target cells). The assays were conducted in gelatin media and cell viability was measured beginning and end of the assays. The results give credence to conclude that (i) TBT affect NK cell function even at 1 h preincubation at 200 nM exposure concentration. (ii) Interleukin2 (IL2) does not consistently protect the suppressive effect of TBT under *in vitro* experimental conditions.(iii) 24 h exposure to 200 nM TBT decreases the NK cell binding to tumor cells.

PHA037 Diethylnitrosamine-induced DNA Adducts in the Medaka Small Fish Model. Law, J.M.* and Marquart, M., North Carolina State University, Raleigh, NC. Small fish models are being used with increasing frequency for carcinogenicity testing and comparative cancer research in the U.S., Canada, and Europe. However, there is a need to further define the early biochemical events of carcinogenesis in these species. Identification and quantitation of DNA adducts can integrate all of the various factors involved in chemical exposure, uptake, distribution, and biotransformation of a putative carcinogen. In the present study, Japanese medaka (*Oryzias latipes*) were exposed to the alkylating agent, diethylnitrosamine (DEN), in the ambient water. Liver DNA was analyzed for O^6-ethylguanine (O6EG), O^4-ethylthymidine (O4ET), and O^2-ethylthymidine (O2ET) by the immuno-slot-blot technique and by LC/mass spectrometry. While fish exposed to 10 ppm DEN had liver DNA adduct levels at or only slightly higher than background levels, those exposed to 100 ppm averaged 34 and 53 pmol O6EG/μmol guanine, 15 and 41 pmol O2ET/μmol thymidine, and 2 and 6 pmol O4ET/μmol thymidine at 0 and 24 hours post exposure, respectively. The results of this study show that, under these short-term exposure conditions, ethyl-DNA adducts appear to accumulate in medaka liver tissue in a sublinear (i.e., nonlinear) fashion after aqueous exposure to DEN. Thus, critical DNA repair enzymes such as O^6-alkylguanine DNA alkyltransferase, relatively efficient at lower carcinogen levels, are probably saturated at the 100 ppm concentration level of DEN.

PHA038 Development of a Sensitive Antibody-based Method to Detect UV-induced DNA Damage in Larval Fish. Iamonte, T.N.* and Bradley B.P, University of Maryland, Baltimore County, Baltimore, MD, Mori T., Nara Medical University, Kashihara, Nara 634, Japan. The release of manufactured chlorine and bromine containing compounds has resulted in a thinning of the stratospheric ozone layer. An important consequence of this atmospheric change is an increase in solar radiation, particularly the more biologically active shorter wavelengths, reaching the Earth's surface. Shorter wavelength radiation (UV-B, 290-320 nm) can penetrate clear water 40 m or more. Larval fishes and other plankton drifting in the first few meters of the water column are predicted to suffer UV damage. The main forms of UV-B induced DNA damage are cyclobutane pyrimidine dimers (CPDs) A sensitive enzyme linked immunosorbent assay (ELISA) to detect CPDs in the UV-B irradiated DNA of larval fishes has been developed. The monoclonal antibody TDM-2 to CPDs was used as a primary antibody. Signal amplification was carried out using a biotin-streptavidin system with 0-phenylene diamine as the color substrate. Absorbance values were measured at 490 nm. The ELISA protocol, examples illustrating detection of DNA damage in larval medaka exposed to artificial UV-B radiation and discussion relevant to the method are presented.

PHA039 Diclofenac Sodium - Induced Mutagenecity in Mouse *Mus musculus* Bone Morrow Cells. M. Krishnamoorthy*, S. Laheri, and M.A. Rahiman, Mangalore University, Mangalagangothri, India. The potent non-steroidal anti-inflammatory drug Diclofenac sodium is chemically sodium [2, 2,6-dichloroanilino-phenyl)] acetate used in the treatment of rheumatic arthritis. The genotoxic potential of it was assessed in the mitotic metaphase chromosomes of swiss albino mouse *Mus musculus*. Three different doses viz. 2.5, 5.0, and 10.0 mg/kg body weight were injected intraperitoneally. 2.5 mg/kg corresponds to human therapeutic dose. Five mice/dose were sacrificed at 12, 24, and 48 hr. Interval to study the dose- and time-yield effect. Distilled water, placebo and 50 mg/kg anti-cancer drug cyclophosphamide administered positive control mice were similarly sacrificed. Bone marrow mitotic chromosome preparations were made by Colchicine -hypotonic- flame dry-buffered Giemsa staining technique. 100 well spread metaphase stages per mouse were screened for structural and numerical chromosome anomalies like break, gap, ploidy, centromeric association, translocation, stickiness, fragmentation, etc. 2000 cells per animal were scored for mitotic index (MI) value. Data were subjected to 't' test. The % age of individual type of aberrations like gap, fragmentation and stickiness were only significant in the positive control mice. The total aberration %age was significant at doses of 5.0 and 10.0 mg/kg of Diclofenac and in the positive mutagen cyclophosphamide at all time intervals. No significant different in the MI value was observed. Hence, Diclofenac was found to be mutagenic only above the human therapeutic dose in the mouse test system.

PHA040 Nickel subsulfide can protect human fibroblasts from the mutagenic effects of benzo[a]pyrene diolepoxide. Hamdan, S., Morse, B., and Reinhold, D., Western Michigan University, Kalamazoo, MI. We have previously reported that, under certain conditions, potassium dichromate can protect human fibroblasts from the mutagenic effects of benzo[a]pyrene diolepoxide (BPDE) (Tesfai, et al., Mutation Res., in press). Nickel subsulfide was used here in place of potassium dichromate to determine if this effect was specific for chromium, or possibly a general effect of transition metals. Normal human fibroblasts were preincubated for 46 hours with 25 mM nickel subsulfide followed by a 2 hour incubation with both 25 mM nickel subsulfide and 0.30 mM BPDE. Control experiments with either no carcinogen or only nickel subsulfide or BPDE were included. The combined carcinogen treatment showed an additive effect on the relative cloning ability of the cells when compared to cells treated with either nickel subsulfide or BPDE. The mutant frequency at the HPRT gene, however, showed that cells treated with both carcinogens had a significant reduction in mutant frequency when compared to cells treated only with BPDE. This protective effect was dependent upon the 46 hour preincubation with nickel subsulfide since the effect was not seen when cells were simply treated for 2 hours with both carcinogens. This antagonistic effect was also shown to be dependent upon the concentration of nickel subsulfide used since a decreased effect was observed at 5.0 mM nickel subsulfide, and almost no effect was observed at 1.0 mM. The antagonistic effect on mutant frequency appeared to be species specific since it was not observed when rat fibroblasts replaced normal human fibroblasts in the protocol described above. These results, along with our previous work, suggest that transition metals can reduce the mutagenic effects of BPDE, and this effect may be species specific.

PHA041 **Subcellular Copper Distribution in Gills of *Corbicula fluminea* after Short-term Exposure.** Oliver, C.; Pilloff, M.; Belaich, M. & Porta A*. Laboratorio de Ecotoxicología y Química Ambiental, Departamento de Ciencia y Tecnología, Universidad Nacional de Quilmes. A known response to heavy metals (as Cd (II), Cu (II), Hg (II)) is the induction of well known metal-binding proteins as metallothioneins. These proteins are small, cystein rich and they can bind efficently heavy metals. This response has proved to be very useful for metal contamination assesment. In this sense, metallothionein mussels has been applied in biomonitoring programs as FAO. Marine mussels as *Mytilus edulis* show induction of metallothioneins by cadmium and copper after 48 hours of exposure, while freshwater mollusks slowly induce them after fifteen days at least. In order to understand better this differential response, copper incorporation and metallothionein-like proteins into the gills of freshwater bivalve *C. fluminea* were studied. Specimens were exposed to Cu(II) 0.50 mg L^{-1} for twelve hours, control and exposed clams were sampled each 90 minutes. Copper was measured at two levels, cytosolic soluble fraction and pellet after centrifugation. Cytosolic metallothionein-like proteins were analyzed by gel electrophoresis and copper was measured using an atomic absorption spectrophotometer. A significant increase of the observed cytosolic copper concentration at five hours seems to be transferred later to pellet fraction (with a maximum about 6 hours). Variation of metallothionein-like proteins and copper level were not observed in control samples along of the experiment. Slightly changes of metallothionein-like protein level were observed on bivalves exposed to copper. High copper uptake was observed during the experience, suggesting that bioavailability of metal is not the unique explanation of poor induction of metallothionein.

PHA042 **Influence of Previous Exposure to Heavy Metals on Tolerance in *Baetis tricaudatus* (Ephemeroptera: Baetidae).** E. Harrahy*, W. Clements, N. DuTeau, W. Black, and B. Beaty, Colorado State University, Fort Collins, CO. The purpose of this research is to compare tolerance to heavy metals and genetic variability among populations of the mayfly, *Baetis tricaudatus* (Ephemperoptera: Baetidae). To test the hypothesis that previously exposed populations are more tolerant to a mixture of heavy metals, a series of laboratory toxicity tests were conducted. *B. tricaudatus* were collected from stations located above and below a U.S. EPA Superfund site contaminated with metals on the Arkansas River, and then exposed in the laboratory to a mixture of cadmium, copper, and zinc for 4, 7, or 11 days. Results indicate significant differences in mortality among both populations and treatments, with previously exposed mayflies more tolerant to the subsequent exposure to metals. Bioaccumulation also differed among populations. However, while growth differed among treatments, it did not differ among populations. To test the hypothesis that previously exposed populations have less genetic variability than naive populations, we are conducting a population genetic study using a phylogenetic approach. Fifty *B. tricaudatus* were collected from each of eleven sites on the Arkansas River (five located above the U.S. EPA Superfund site, and six below), four sites on Chalk Creek (tributary to the Arkansas River), and three sites on the Cache la Poudre River (as an outside control), at each of three time points. Due to inconsistent amplification of our initial choice of the12S mitochondrial DNA gene, we designed species-specific primers for the ND1 gene. The frequency of individual haplotypes is being determined and compared for sites located above and below the U.S. EPA Superfund site. Preliminary results indicate decreased genetic variability downstream. This narrowing of genetic diversity, through elimination of sensitive genotypes and retention of resistant genotypes, may result in increased susceptibility of these populations to further stress.

PHA043 **Role of adsorption in the *in vitro* toxicity of fluoranthene.** Schirmer, K*, Chan, AGJ, Bols, NC, University of Waterloo, Waterloo, ON, Canada. One characteristic of hydrophobic contaminants is their high affinity to surfaces, including those of tissue culture devices. For example, fluoranthene was found to adhere to tissue culture plasticware with high affinity. Therefore, we addressed the question whether adsorbed fluoranthene can elicit cyto- and photocyto-toxic responses in the rainbow trout gill cell line, RTgill-W1. Dissolved fluoranthene (297 nM) was added to wells of 48-well tissue culture plates and allowed to adsorb for 48 h before the solution was removed and wells air dried. Cells were plated onto the fluoranthene-coated culture surface, and the attachment process was monitored for up to 24 h with the esterase substrate, CFDA-AM. Interestingly, cell attachment was slightly enhanced in the presence of fluoranthene. A subsequent 2 h UV-irradiation diminished within 24 h the ability of cells to convert and retain the fluorescent product of CFDA-AM and to reduce the alamar Blue indicator dye by more than 70%. These results suggested that photocytotoxicity is due to the cells acquiring fluoranthene from the growth surface and/or their close proximity to the fluoranthene coated surface. The ability of cells to specifically desorb fluoranthene from the growth surface was tested by allowing cells to attach to a culture flask that previously had been exposed to fluoranthene (297 nM), and to subsequently UV-irradiate these cells in the absence of adsorbed or dissolved fluoranthene. Again, photocytotoxicity was observed 24 h later but was much less than for the cells that were UV irradiated in the presence of adsorbed fluoranthene. Thus, although cells can acquire sufficient fluoranthene from the growth surface to be sensitized to a subsequent UV irradiation, adsorbed fluoranthene in their close proximity can greatly enhance this effect.

PHA044 **2,3,7,8-Tetrachlorodibenzo-*p*-dioxin Alters Cardiovascular and Craniofacial Development and Function in Sac Fry of Rainbow Trout (*Oncorhynchus mykiss*).** M.W.* and Peterson, R.E., University of Wisconsin, Madison, WI; Spitsbergen, orodibenzo*p*-dioxin (TCDD) toxicity in rainbow trout sac fry, are yolk sac edema, hemorrhage, craniofacial malformation, and growth retardation culminating in mortality. Our objective was to determine the role of cardiovascular dysfunction in the development of this toxicity. An embryotoxic TCDD dose (385 pg/g egg) caused a progressive reduction in blood flow in rainbow trout sac fry manifested first and most dramatically in the 1^{st} and 2^{nd} branchial arches and vessels perfusing the lower jaw. Blood flow was reduced later in the infraorbital artery and occipital vein of the head as well as segmental vessels and caudal vein of the trunk. Reduced perfusion occurred last in gill branchial arteries involved with oxygen uptake and the subintestinal vein and vitelline vein involved with nutrient uptake. Although heart rate throughout sac fry development was not affected, heart size at 50 days post-fertilization (l development was arrested near hatch giving rise to craniofacial malformations in which the jaws and anterior nasal structures were underdeveloped. Unlike the medaka embryo where TCDD causes apoptosis in the medial yolk vein, endothelial cell death was not observed in rainbow trout sac fry. These findings suggest a primary role for arrested heart development and reduced perfusion of tissues with blood in the early life stage toxicity of TCDD in trout.

PHA045 **Dioxin-like Embryotoxicity of Lake Michigan Lake Trout Extract to Developing Lake Trout.** Wright, P.J.*, Environmental and Contaminants Research Center, Columbia, MO; Tillitt, D.E., Environmental and Contaminants Research Center, Columbia, MO. Planar halogenated hydrocarbons (PHHs) are known to be present in the Great Lakes ecosystem in sufficient concentrations to cause adverse effects on reproduction in certain species of fish. Previously, we demonstrated the toxicity of a PHH mixture through the nanoinjection of environmental extracts into newly fertilized eggs from two strains of rainbow trout. The current study was designed to investigate the embryotoxicity of a complex organic extract on developing lake trout embryos. Graded doses of the final extract were injected into eggs of hatchery reared lake trout. Total TEQs in the Lake Michigan lake trout sample were 14.7 pg TEQ/g. The extract of the Lake Michigan lake trout was embryotoxic to lake trout embryos in the laboratory with an LD50 value of 7 eggEQ in lake trout. The LD50 of the Lake Michigan extract in terms of TEQs was 103 TEQs/g of lake trout egg. The estimated ED50 values for sublethal responses were: 7.4 eggEQ for craniofacial anomalies, 3.0 eggEQ for yolk sac edema (YSE), and 0.4 eggEQ for hemorrhage in the lake trout embryos. The LD50 of TCDD in lake trout was 81 pg/g egg and was similar to previous reports. Our previous studies with rainbow trout embryos injected with this same extract predicted an LD50 in lake trout of 4 eggEQ based on the Erwin strain of rainbow trout or 7 eggEQ based on the Arlee strain of rainbow trout. This study confirms our earlier conclusions that PHHs in Lake Michigan lake trout are above a threshold for adverse sublethal effects and these compounds may have implications on the lack of recruitment in certain Great Lakes lake trout populations.

PHA046 Early Life Stage Toxicity of 2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) and PCB 126 to Bull Trout. Lawonn, M., Loeffler, I.K., Andreasen, E., and Peterson, R.E., University of Wisconsin, Madison, WI; Fredenberg, W., U.S. Fish and Wildlife Service, Creston, MT; Cook, P.M., U.S. EPA, Duluth, MN. Population declines of bull trout, Salvelinus confluentus, have occurred in recent decades throughout much of the species' natural range in the Columbia River watershed and other river systems in western Canada. Bull trout in the U.S. are now listed as threatened under the Endangered Species Act. Rehabilitation efforts are concerned with habitat loss and fragmentation, water quality degradation, and competition with non-native species. Virtually no information exists regarding the vulnerability of bull trout to environmental contaminants. The lake trout, Salvelinus namaycush, is closely related to the bull trout and is the most sensitive species known for early life stage mortality associated with exposure of embryos to TCDD and related AhR agonists. In order to determine the sensitivity of bull trout relative to lake trout, eyed embryos were exposed to graded concentrations of [^{3}H]-TCDD and PCB 126 in water. Highest concentrations of both chemicals in eggs resulted in accelerated hatching and embryo mortality. Subsequent sac fry mortalities were associated with signs of toxicity similar to those observed in other trout species (yolk sac and pericardial edema; periocular and yolk sac hemorrhage; craniofacial malformations; and exopthalmia). Reduced blood flow was observed in sac fry with signs of toxicity. Preliminary examination of dose-dependent mortality data indicates that bull trout are approximately two times more sensitive to TCDD than lake trout based on comparison of eyed embryo exposures for the two species. A toxicity equivalence factor (TEF) for PCB 126 will be determined. Survival and development of the remaining bull trout fry during the first two months of feeding are being evaluated. Final results from this study will enable screening level assessments of risks for early life stage mortality and consequent recruitment failure in bull trout populations on the basis of estimated or measured concentrations of AhR agonists in eggs.

PHA047 Dioxin toxicity in lake trout embryos with low thiamine levels. Wright, P.J.*; Tillitt, D.E.; Zajicek, J., Environmental and Contaminants Research Center, Columbia, MO; Anderson, M., Hagler-Bailley Consulting Inc., Boulder, CO; Fitzsimons, J., Great Lakes Laboratory for Fisheries and Aquatic Sciences, Burlington, Ontario, Canada; Holey, M., U.S.F.W.S., Green Bay, WI; Honeyfield, D.C., U.S.G.S., Wellsboro, PA. An interaction of nutritional and contaminant factors may be occurring in Great Lakes salmonids affected by Early Mortality Syndrome (EMS). Adult female salmonids in the Great Lakes accumulate dioxin-like compounds via contaminated prey, water and sediments. Coinciding with this, adult salmonids feed on diets which are either thiamine deficient, or contain high quantities of thiaminase-containing prey. Although the degree of thiamine deficiency is not great enough to directly affect the overall health of adult fish, females, particularly those from Lake Michigan and Lake Ontario, spawn eggs which are deficient in thiamine. Pharmacological doses of thiamine ameliorated certain adverse effects of TCDD in rainbow trout and Japanese medaka embryos. The suggestion of these results was that an interaction exists between thiamine and dioxin-induced embryo toxicity. The studies reported here were designed to reflect more environmentally-relevant interaction among these stressors; notably low thiamine content in lake trout eggs and elevated dioxin. Lake trout eggs from both feral and laboratory sources were spawned, and then exposed to 2,3,7,8-TCDD and/or a complex extract of Great Lakes walleye via injection or waterbath treatment. Thiamine supplementation of low thiamine eggs prior to contaminant dosing reduced mortality at doses that were less than 100% lethal, and also had significant effects on the occurrence of gross lesions in exposed lake trout fry. These results indicate that thiamine deficiency may potentiate the effects caused by dioxin exposure on egg and fry growth and survival.

PHA048 Acute Toxicity of Oganotins and Sediment Extracts to Fertilization and Development of Sea urchin, Anthocidaris crassispina. Shim, W.J.*, Aminin, D., Agafonova, I., and S.H. Lee, Korea Ocean R & D Institute, Korea. Comparative toxicities of eight organotin compounds (tributyltin, dibutyltin, monobutyltin, triphenyltin, diphenyltin, monophenyltin, trimethyltin, and dimethyltin) were evaluated with sea urchin, Anthocidaris crassispina. ED50 and threshold concentrations of each organotin compounds were determined with four separate tests: 1) 8-blastomer development, 2) treated sperm fertilization, 3) treated egg fertilization, 4) pluteus larvae mortality. Among the four tests, the fertilization test with organotin treated sperms was the most sensitive. Tributyltin (TBT) was the most toxic among eight organotin compounds with all tests. The lowest ED50 for TBT was 0.06 mg/ml in treated sperm test, whereas dimethyltin showed the highest ED50 (29 mg/ml). The lowest threshold concentration of TBT was <0.0017 mg/ml in larvae mortality test. In three butyl-, phenyl-, and methyltin groups, triorganotin compounds were the highest toxic, respectively. The methylene chloride extracts from surface sediments of the major harbors were tested with 8-blastomer development. ED50 of sediment extract showed significant correlation with total butyltin concentrations of the sediment excluding the sample which had exceptionally high (13,000 ng/g) butyltin levels.

PHA049 Uptake efficiencies of chemicals across the fish gill. Koyama, J.* and Ikeda, K., National Research Institute of Fisheries Science, Kanagawa, Japan. Uptake efficiencies of chemicals across the fish gill, which relate to bioconcentration of chemicals and are one of the important parameter in physiologically based toxicokinetic model, were determined in vivo using marine fish, red sea bream. The water were taken from oral and opercular cavity of intact fish in respirometer-metabolism chamber and the concentrations of chemicals in the water were determined. Uptake efficiencies of chemicals were calculated with the differences of the concentrations between oral and opercular cavity water. Uptake efficiencies of each chemicals exhibited relatively constant and mean uptake efficiencies of fenitrothion, tributyltin oxide, triphenyltin chloride and naphthalene were 49, 24, 21 and 70%, respectively. The linear relationship was observed among their mean uptake efficiencies and Kows. On the other hand, uptake efficiency of cadmium, one of the hazardous heavy metals, varied between 0.7 to 13%. Uptake efficiencies of cadmium at lower concentration, 0.01mg/l, exhibited relatively higher value than those at higher concentration, 1.1mg/l. These results suggest that the uptake mechanisms of heavy metals are different from organic chemicals.

PHA050 Relative Toxicity of Polychlorinated Naphthalenes (PCNs) to Medaka After *In ovo* Exposure: Early and Partial Life Cycle Assessments. Pastva, S.D.*, Villalobos, S.A.*, Blankenship, A.L., Kannan, K., Giesy, J.P., Michigan State University, East Lansing, MI; Papoulias D.M., Meadows, J., Tillitt, D.E., USGS Environmental and Contaminants Research Center, Columbia, MO. PCNs have similar properties and uses as some PCBs, and have been detected in biota. The toxicity of several PCN mixtures (Halowax) was evaluated in medaka (*Oryzias latipes*), using the embryo nanoinjection method. Medaka eggs (early gastrula) were injected with 0.5 nl of triolein (vehicle control) or 0.5 nl of 4-5 graded doses (0.3-30 ng/embryo) of halowax 1014, halowax 1013, or halowax 1051. Following exposure, embryos were raised and fry reared for approximately 4 months, at which time fish were killed (after sexual maturity). Responses were evaluated as early life cycle (ELC), and partial life cycle (PLC) assessments. For ELC, lethality and sublethal alterations (e.g., craniofacial, cardiovascular, myoskeletal, abnormal/delayed hatch) were monitored for the first 9 days, and a severity index computed. PLC assessment examined: survival/mortality until adulthood, gonadosomatic index (GSI), and morphometry. Results showed halowax 1014 as the most toxic (LD50 4.5 ug/g embryo) mixture, while halowax 1013 or 1051 were significantly less toxic. GSI was significantly decreased in halowaxes 1014 and 1051 treated females. When compared to dioxin's LD50 for nanoinjected embryos (0.8 ng/g embryos), halowax 1014 was 5700x less potent. ELC assessments were as good of a predictor as PLC assessment. However, when compared to the HII4E-Luc cell assay for Ah receptor interaction, the order of relative potencies in medaka did not behave as predicted.

PHA051 Occurrence of PCBs and Organochlorine Pesticides in Tissues of Stripped Weakfish (Cynoscion striatus) from Blanca Bay, Argentina. Lanfranchi, A.L.*, CONICET-UNMP, Mar del Plata; Aizpún de Moreno, J.E., UNMP, Mar del Plata; Moreno, V.J., UNMP, Mar del Plata; Janiot, L.J., Servicio de Hidrografía Naval, Capital Federal; Menone, M.L., CONICET-UNMP, Mar del Plata, Argentina. The concentration of Polichlorinated Biphenyls (PCBs) and Organochlorinated Pesticides (OCPs) were determinated in liver, muscle and reproductive organs from male and female stripped weakfish (*Cynoscion striatus*), caught during post spawning stage in Blanca Bay, Argentina, in April 1997. Concentrations of PCBs and OCPs were determined by GC-ECD. These levels were reported on a wet weight basis and normalized to lipid content. The highest concentration of organochlorine compounds were found in male and female liver, but a large concentration was also detected in ovary. The concentrations of OCPs in the livers and gonads of both sexes was in the order: DDT and metabolites > total cyclodienes > total HCHs, while in the ovaries was: total cyclodienes > total HCHs > DDT and metabolites. In all tissues, the predominant PCBs were Tri, Tetra, Penta and Hexachlorobiphenyl congeners, with large contribution from the highly persistent congeners 138 and 153. The concentrations of PCBs and OCPs in the ovary were below the levels considered safe for normal reproduction in other fish species.

PHA052 Relationship between PCB Concentrations and Reproduction in the Silverside *Odontesthes bonariensis,* from Mar Chiquita Coastal Lagoon, Argentina. Menone, M.L. *, CONICET-UNMP, Mar del Plata; Aizpún de Moreno, J.E., UNMP, Mar del Plata; Moreno, V.J., UNMP, Mar del Plata; Metcalfe, T., Trent University, Canada; Metcalfe, C.D., Trent University, Canada; Lanfranchi, A.L., CONICET-UNMP, Mar del Plata, Argentina. Changes in the wet weight, lipid and PCB levels of muscle, liver, gonad, gut and mesenteric fat of spring spawning silverside, of both sexes from Mar Chiquita lagoon, were examined in relation to stage of sexual maturation from August (gonadal development noticeable) until December (resting stage). PCBs were determined by GC-ECD. In stage II (August) mesenteric fat was absent and lipid levels of liver and gonads were low. Great quantities of PCB could be released into the blood stream when mesenteric fat is utilized. The fate of these lipophilic compounds could be gonads and liver, in which relatively high PCB levels were found in both sexes. Mesenteric fat, which acts as a tramp of PCB (until 148 ppb), in males and females (stages III-VII) was present. Simultaneously, wet weight and lipid levels of liver and gonads were highly increased. Negative correlations between lipid burden and PCB levels were observed. During spawning, large quantities of PCBs left the fish and their levels fell in ovaries. In resting stage both lipid and PCB levels increased in all tissues. The patterns of mesenteric fat and muscle were dominated by penta and hexachlorobiphenyls with large contributions from the highly persistent congeners PCB 118, 138 and 153. During maturation period, the gonads showed a not clear pattern of PCBs distribution (dominated by three, tetra, penta and hexabiphenyls congeners), but in resting period the composition was similar to those of mesenteric fat and gut in both sexes.

PHA053 Interlaboratory Comparison of a 96-Hour *Mysidopsis bahia* Bioassay using a Water Insoluble Synthetic Based Drilling Fluid. Rabke, S.P.*, M-I *L.L.C.*, Houston, TX, Candler, J.E., M-I *L.L.C.*, Houston, TX. A 96-hour bioassay using the mysid *Mysidopsis bahia* is the current compliance test for drilling fluids discharged offshore in the U.S.A. Previous round robin studies of this test has focused on water based drilling fluid (WBF) formulations. Synthetic based drilling fluids (SBF), which are not water soluble, have not been subjected to the same type of round robin testing. An interlaboratory comparison (round robin) was conducted to evaluate the test method. The SBF was mixed in 20 ppt seawater at a ratio of 1:9. The resulting mixture was stirred for 5 minutes and allowed to settle for one hour. The suspended particulate phase was decanted and used as the test material. The *M. bahia* were exposed to the test material in a 96-hour static non-renewal toxicity test. The end point of the test was an LC_{50}. Six different facilities were included in the round robin study. Observations of the methodologies used were made for each facility. Results of the round robin study on SBFs are compared with previous round robin testing with SBFs. In addition to reporting statistical analysis of the results, such as coefficient of variation and standard deviations, observations of techniques used to set up the tests are reviewed to identify any differences in lab technique that may have affected the results.

PHA054 The Sensitivity of Grass Shrimp, *Palaemonetes pugio,* Embryos to Organophosphate-Induced Acetylcholinesterase Inhibition. Lund, S.A.*, Fulton, M.H., Key, P.B., National Ocean Service, Charleston, SC. A common inhabitant of salt marshes along the Atlantic and Gulf coasts of North America is the grass shrimp, *Palaemonetes pugio.* Within these salt marshes, grass shrimp fulfill a vital role in energy transfer and ecosystem stability. In South Carolina, agricultural fields are often located adjacent to salt marshes. Since the reproductive period in these animals corresponds to the peak agricultural growing season, grass shrimp at all developmental stages are at risk for pesticide exposure. Organophosphorus compounds (OPs) are widely used as agricultural insecticides. The toxic action of OPs results from the inactivation of acetyicholinesterase (AChE). Although adult and larval *P. pugio* have been widely used in toxicity tests to examine various effects of OPs, little research has focused on the effects of these compounds in embryos. The goals of this study were to examine the development of AChE activity in grass shrimp embryos, assess their sensitivity to OP-induced AChE inhibition, and to compare the sensitivity of the embryos to that of larval and adult *P. pugio.* Quantifiable AChE activity first appeared in Stage V embryos and increased as development proceeded. Chlorpyrifos 24-h EC_{50}s were 0.49 µg/L for Stage VI embryos and 0.33 µg/L for Stage VII embryos, while 24-h malathion EC_{50}s were 29.93 µg/L for Stage VI embryos and 55.53 µg/L for Stage VII embryos. Embryonic *P. pugio* were as sensitive to chlorpyrifos and malathion as larval and adult organisms. These findings suggest that *P. pugio* embryo bioassays may provide reasonable predictions as to how these contaminants will affect both larval and adult *P. pugio.* Future research will examine the relationship between OP-induced enzyme suppression and acute toxicity in embryonic grass shrimp.

PHA055 Animal and Cell Culture Assays for Modeling the Toxicity of Azadirachtin Insecticides. Goktepe, I. and Plhak, L.C., Department of Food Science, Louisiana State University, Baton Rouge, LA. Azadirachtin (AZA) insecticides are marketed as natural insecticides having anti-feedant and anti-molting effects on arthropods. However, the increasing popularity of these insecticides may potentially cause ecological risks to aquatic crustaceans and mollusks. The objectives of this study were 1) to evaluate the acute toxicity of AZA-insecticides, namely Neemix™ and Bioneem™ on crayfish, freshwater snails, blue crab, oyster, and *Daphnia* and compare their toxicity to that of pure-AZA and 2) to develop an *in vitro* system to asses the toxicity of pure-AZA and AZA-insecticides on cell cultures. A 96 h acute toxicity test was conducted on all species named above at concentrations from 0 to 20 ppm AZA for Neemix™ and Bioneem™, 0 to 30 ppm for pure-AZA. Hybridoma cultures were exposed to pure-AZA, Neemix™ and Bioneem™ at concentrations of 0, 0.001, 0.1, 1, 10, and 100 ppm AZA for 24, 48, 72, and 96 h. A colorimetric assay based on the reduction of MTT tetrazolium dye by active mitochondria was used to evaluate the toxicity. The sensitivity of animals to both Neemix™ and Bioneem™ was in the following order, *Daphnia*>oyster>blue crab>freswater snails>crayfish with LC_{50} values of 0.0202>0.25>1.16>2.45>6 ppm AZA, respectively. Pure-AZA did not show significant toxicity to crayfish and freshwater snails at the tested concentrations, but LC_{50} of pure-AZA for *Daphnia* was 0.246 ppm AZA. It was also observed that at higher concentrations of insecticides, the number of molted crayfish decreased by approximately 33%. At concentrations 10 and 100 ppm AZA, Neemix™ and Bioneem™ were toxic to hybridoma cells, whereas pure-AZA was not toxic to hybridoma cells at all concentrations used. Modeling of the toxicity of azadirachtin insecticides indicate that Neemix™ and Bioneem™ were more toxic than pure-AZA at equivalent concentrations which suggest that they contain additional toxic compounds.

PHA056 Measures of Behavioral Alterations in Rainbow Trout *(Oncorhynchus mykiss)* Resulting from Acute Exposure to Insecticides. Brewer, S. K.*, DeLonay, A. J., Beauvais, S. B., Little, E. E., and Jones, S. B. USGS-BRD, Environmental and Contaminants Research Center, Columbia, MO, USA. Behavioral responses are affected by sublethal concentrations of toxic chemicals. Abnormal behavioral responses reflect changes in the nervous system such as activity of neurotransmitter pathways, sensory perception, or motor activity. Such alterations may reduce growth, survival, and fitness, and eventually create undesired population effects. We correlated behavioral responses with physiological endpoints as a way to link behavior with underlying physiology. Rainbow trout (RBT) fingerlings were exposed to the organophosphorous (OP) insecticides, diazinon and malathion, and the pyrethroid, fenvalerate, in static renewal experiments. Video samples were obtained for behavioral analysis following 24-or 96-h exposures and a 48-h recovery period. We measured four locomotory behaviors (distance traveled, speed, tortuosity of path, and rate of change in direction) and three physiological endpoints (cholinesterase inhibition, binding of muscarinic cholinergic receptors. The behavioral endpoints were effective in demonstrating significant trends of contaminant effect over duration of exposure. Behavioral and physiological responses of fish were similar although behavior was a more integrative endpoint for the chemicals we used. Correlations between swimming and ChE varied depending on whether the pesticides were cholinesterase inhibitors. These results demonstrate the applicability of behavioral endpoints in toxicity assessments.

PHA057 Physiological and Behavioral Measures of Neurotoxicity in Rainbow Trout, ***Oncorhynchus mykiss.*** Beauvais, S.L.*, Brewer, S. K., Jones, S.B., Little, E.E. USGS/BRD), Environmental and Contaminants Research Center, Columbia, MO. Neurotoxicant exposures can alter physiological functions, resulting in changed behavior. For example, organophosphate insecticides (OPs) inhibit acetylcholinesterase, causing increased concentration of the neurotransmitter, acetylcholine, with subsequent overstimulation of cholinergic pathways and abnormal behavior. We investigated correlations between changes in physiological variables and resulting behavioral responses in larval rainbow trout (RBT) exposed to sublethal concentrations of neurotoxicants. RBT were exposed to the OPs, diazinon and malathion, and the pyrethroid, fenvalerate, in static renewal experiments. Treatments consisted of three or four concentrations of each chemical and three exposure durations, 24 h, 96 h and a 48-h recovery period following a 96-h exposure. RBT were videotaped individually to assess locomotory behaviors. Brain tissue from these fish was then used to measure the physiological endpoints, cholinesterase (ChE) activity and muscarinic cholinergic receptor (MChR) binding. ChE activity was significantly inhibited by exposure to both diazinon and malathion, though not by exposure to fenvalerate. There was no significant effect on MChR number or affinity by exposure to the OPs, but there was a concentration-dependent trend of a decrease in receptor number. In RBT exposed to fenvalerate, there were no significant differences in MChR number or affinity at both 24 and 96 h, but fish in the 48-h recovery group showed a significant increase in receptor number. There were significant correlations between ChE activity and swimming speed for both OP exposures, though not for fenvalerate, which did not cause ChE inhibition. These results suggest that correlations between physiological and behavioral changes previously seen in mammals also occur in fish.

PHA058 Evaluation of the Japanese Medaka ***(Oryzias latipes)*** **Embryo Larval Assay to Assess the Extractable Organic Phase Toxicity of Contaminated Sediments.** Owens, K.D.* and Baer, K.N., Northeast Louisiana University, Monroe, LA. Over the past few decades, industrial and agricultural processes have become major sources of contamination in the environment, particularly aquatic ecosystems. The early life stages of teleost species are generally the most sensitive to environmental pollutants. One method currently available for measuring embryo toxicity is the Japanese Medaka (*Oryzias latipes*) Embryo Larval Assay (MELA). The objective of this study was to evaluate the MELA as a screen for developmental toxicity of contaminated sediments. Previous studies show elevated concentrations of organochlorines, primarily ΣDDT and toxaphene, in sediments and resident fish in the Tensas River Basin, Louisiana. The Tensas River Basin is located within the Mississippi River alluvial floodplain of northeast Louisiana. Although toxaphene and ΣDDT were found to be ubiquitous throughout the study area, there appeared to be a north-south gradient for the Tensas River mainstream. The highest sediment concentrations of ΣDDT occurred in the headwaters of the Tensas River (geometric mean, 0.125 ug/g), but toxaphene apparently was below detection limits. Channel catfish sampled from the headwaters of the river contained whole body fish residues of toxaphene and ΣDDT as high as 11.0 and 3.76 ug/g, respectively. It is unclear whether resident fish are exhibiting toxicity, such as reproductive or developmental impairment, due to this contamination. In view of these considerations, sediment samples from selected sites along the Tensas River (high, medium, and low sites of contamination) were extracted using hexane: acetone (1:1) and exchanged to DMSO. These extracts were topically applied to medaka embryos (48 hours old) and observed for developmental toxicity. The MELA appears promising as a sensitive indicator of developmental toxicity from contaminated sediments.

PHA059 Investigation of Temperature Effects on Toxicity of Organophosphate Insecticides to ***Chironomus tentans*** **Using a Body Residue Approach.** Ternes, M.A.*, J.B. Belden, and M.J. Lydy. Wichita State University, Wichita, KS. The effects of temperature on the toxicity of chemicals in aquatic systems have been well studied, however few if any studies have examined the temperature effects on whole body residues in organisms. Acute toxicity and whole body residues were determined for selected organophosphate insecticides (OP's) in exposed *Chironomus tentans.* The OP's used in this study included chlorpyrifos, methyl parathion, and malathion. Standard 96 hour toxicity tests were utilized to obtain toxicity data. Body residues were evaluated by use of radiolabeled OP's linked with liquid scintillation counting. Results have been obtained for determination of temperature effects at 10, 20, and 30°C. Initial results indicate increased toxicity with increased temperature. Whole body residue data indicates that the increase in toxicity may be attributable to an increase in uptake of the chemical by the organism due to increased respiration as a result of the increased temperature.

PHA060 The Excessive Danger of Anticholinesterase Compounds in Acidified Surface Water. Tonkopii, V.D.*, Zagrebin, A. O., Institute for Lake Research, Russian Academy of Sciences, St.Petersburg, Russia. Among many xenobiotics entering aqueous media anticholinesterase (antiChE) compounds notable for their high toxicity and selectivity of action are of particular hazard. Many compounds of this class are used as pesticides, drugs and warfare agents. In general these compounds as a rule are nonstable on account of a quick hydrolysis in neutral water pH. With some exception their half-lives are measured by days or weeks but great hazard is that some agents are very toxic especially for hydrobionts and mammals. For study of influence of water acidification on transformation of organophosphorus pesticides the hydrolysis rate of widely use agents (dichlorvos, dipterex, chlorpyrifos, diazonin, malathion, mevinphos, parathion, phosphamidon, TEPP) in different water pH (5.0; 5.5; 6.0; 7.5; 8.5) was determinated. It was observed that in conditions of low pH (5.0 - 6.0) and temperature from 0^0 C to 5^0 C the rate of compounds hydrolysis is very low. The half-lives of agents in these conditions made up 300-1200 days as compared with 15-100 days for neutral pH. The toxicity of organophosphorus compounds in low water pH has been determined in experiments on *Daphnia magna.* Besides the activity of some enzymes (aryl - and carboxylesterase, cytochrome P-450 system) which are hydrolysing the organophosphates was studied with use of *Daphnias* homogenates or microsomes. It was observed that the activity of all enzymes was significantly decreased with increasing of sensitivity of *Daphnias* to the organophosphorus pesticides. Thus it was found that the acidification of freshwater was accompanied by increase of organophosphates stability and their toxicity for *Daphnia magna.* The organophosphorus pesticides in acidified water have excessive danger for humans, animals and particularly for hydrobionts.

PHA061 A Comparison of the daphnids, ***Ceriodaphnia dubia*** **and** ***Daphnia ambigua*** **for their Utilization in Routine Toxicity Testing at the Savannah River Site, Aiken, SC.** Harmon, S.M.*, University of South Carolina, Columbia, SC, Specht, W.L., Westinghouse Savannah River Co., Aiken, SC, Chandler, G.T., University of South Carolina, Columbia, SC. Short-term whole effluent toxicity testing, which is currently a requirement of the U.S. EPA's National Pollution Discharge Elimination System (NPDES), commonly uses the cladoceran species *Ceriodaphnia dubia.* Despite the advantages to using a common test species to model the toxic effects of effluents, it could be argued that toxicity test results would be more meaningful if a wider variety of test organisms were commonly used. One particular argument against *C. dubia* is that tests conducted with this species do not always reflect local, site-specific conditions. The careful selection and use of an indigenous test species would produce a more realistic model of local instream effects and would account for regional differences in water quality. This study addressed the substitution of *C. dubia* with a southeastern U.S. indigenous cladoceran species, *Daphnia ambigua,* for routine regulatory toxicity testing at the Savannah River Site (SRS), a facility operated by the U.S. Department of Energy in Aiken, SC. This investigation determined that *D. ambigua* could be laboratory cultured with only minimal changes to established regulatory protocol and that the life-cycle characteristics of this species were conducive to traditional acute and chronic aquatic toxicity test methods. Acute toxicity tests showed that when comparing LC_{50} values for *C. dubia* and *D. ambigua, D. ambigua* was less sensitive to some toxicants (sodium chloride, copper sulfate, and sodium lauryl sulfate) while more sensitive to others (chlorpyrifos). Results of chronic tests with copper sulfate and sodium chloride resulted in the same NOEC/LOEC values for both species. When exposed to unaltered SRS stream water, *C. dubia* demonstrated a chronic "toxic" response for two of the three streams tested, while reproduction for *D. ambigua* was higher in all stream samples. These results suggest that *D. ambigua* would serve as a suitable replacement for *C. dubia* in regulatory toxicity testing.

PHA062 Summary and Methods Variability Issues of the U.S. EPA Discharge Monitoring Report Quality Assurance Program (DMRQA) Whole Effluent Toxicity Testing (WETT) from 1991 - 1997. Lazorchak, J.M., P.W. Britton, USEPA, Cincinnati, OH, 45268 and J.D. Helm, USEPA, Washington, D.C., 20460. The DMRQA WETT program serves as the primary tool for USEPA and the States to evaluate the quality of the NPDES WETT self-monitoring data. The program evaluates permittees' ability to produce and report accurate toxicity data. Participation of designated major and minor permittees in the DMRQA WETT program is required, based on the authority given to the U.S. EPA under Section 308(a) of the Clean Water Act. The DMRQA WETT program also provides a national consistency and equivalency check on NPDES laboratories producing toxicity data. DMRQA WETT allows EPA to document that equivalent results can be obtained across the country. For the past 7 years the USEPA has been evaluating the laboratories that have been conducting freshwater and marine toxicity tests for specifically designated NPDES discharges. The number of methods and endpoints that were evaluated over this time ranged from 9 methods and 19 endpoints to 31 methods and 47 endpoints. During these 7 studies three different reference toxicants have been used for both freshwater and marine tests. The results from the 7 studies (studies 11-17) can be used to evaluate the interlab variability in the WETT methods. As an example of the analyses of these results: for studies 16 and 17 statistical evaluations (regression analyses) made on acute toxicity tests with fathead minnows (method codes 11, 13, & 14) showed that there were no statistically significant differences between method codes 11 and 13. There were statistical significant differences between method codes 13 and 14 and the endpoint of the three methods were not effected by temperature. LC50 results when using moderately-hard synthetic freshwater or 20 % diluted mineral water were statistically different. Comparisons of results for other method codes and studies will be presented.

PHA063 The Effects of Mixtures of Organophosphorus Insecticides on the Fathead Minnow (*Pimephales promelas*) - Acetylcholinesterase Inhibition. Chappel, M.J.*, Solomon, K.R., Sibley, P.K., Centre for Toxicology, University of Guelph, Canada. Aquatic ecosystems in close proximity to agricultural areas are routinely exposed to mixtures of organophosphorus insecticides. Organophosphorus insecticides are known to cause a depression in acetylcholinesterase (AChE) activity following exposure. For mixtures of organophosphorus insecticides, additive responses may occur within an organism even when all the individual components of the mixture are present at concentrations below their individual thresholds of biological response. Several studies were conducted to determine the effects of both single, binary, and tertiary mixtures (azinphos-methyl, chlorpyrifos, diazinon) of organophosphorus insecticides on an aquatic vertebrate using both mesocosm and laboratory studies. In all studies, brain acetylcholinesterase activity was monitored for up to 7-d. Concentration-response studies indicated good correlation between mortality and brain acetylcholinesterase inhibition and was significantly greater when compared to controls. The concentration-response studies allowed calculation of both individual and mixture LC50's and NOEC's. Using mixture concentrations at 150 x 90th centile of MEC's, inhibition of AChE was up to 90%, while at the 90th centile, inhibition was significantly greater than the controls. Brain acetylcholinesterase inhibition was rapid (within 2-hrs), and is considered a sensitive indicator of toxicity. As an effect measurement, AChE inhibition can be considered a predictive linkage of the assessment measurement of ecological effects to populations of fathead minnows.

PHA064 Toxicological assessment of aquatic samples from Abrolhos National Park - Brazil. Mastroti, R.R.*, University of Sao Paulo, Sao Paulo, SP, Brazil. The Abrolhos National Park was the first Marine National Park established in South America (1983). It includes two coral reef areas and several rocky islands, which are very fragile and complex ecosystems. In spite of representing a crucial part of its Management Plan, no environmental monitoring research has been carried out since the Park creation. In order to assess the environmental impact of constant visitation combined with the presence of a few inhabitants, seven representative sampling sites were selected for examining surface microlayer, water column, and porewater samples. The surface microlayer was sampled using the glass plate method. Samples of the water column were collected using glass flasks which were opened at half depth. The pore water was extracted using plastic syringes connected to air stones previously (~36 h) buried in the sediment. All samples were immediately centrifuged, labeled, and stored at -15±3°C. In order to assess the toxicity of these water samples, the sea urchin embryo test (*Lytechinus variegatus*) was selected due to its sensitivity and because it is easy to perform. Within all samples, only one surface microlayer sample shown significant toxicity. This toxic sample was collected near a boat undergoing repair, thus representing a localized and transient source of pollution. The evaluation of the potential impact resulting from the discharge of domestic wastes may have been biased, however, because during the time of sampling the tide did not reach the discharge points. Only during the highest tides does the discharge directly enter the sea. The results of this preliminary study, however, suggest that the current number of visitors does not represent an environmental hazard to this National Park.

PHA065 Evaluation of the Impact of Climate Change on Waterborne Cryptosporidiosis. Kocagil, P.*, Fisher, A., Shortle, J., The Pennsylvania State University, University Park, PA. Changes in climate driven by greenhouse gas emissions are likely to have a direct impact on the occurrence of waterborne diseases and may result in large economic impacts to society. More frequent or severe meteorological events (e.g. heavy rains, snow melts, swollen rivers) that flush areas of high pathogen concentration into private and public water supplies are likely to increase the risk of disease. Cryptosporidiosis is one of many waterborne diseases whose prevalence is likely to increase with increased precipitation and flooding triggered by climate change. This study evaluates the costs associated with the increased risk of cryptosporidiosis that may occur as a result of climate change. An event tree in conjunction with a cost-of-illness (COI) methodology is used to evaluate the impacts of climate change. Medical treatment costs, the value of time lost from work and leisure, and costs incurred to avoid contamination are included in the COI. The event tree is used to assess vulnerability to health threats from contaminated water. This is accomplished by tracing probabilities of events and projecting risk averting or intensifying actions. This requires substantial information about pathways of contaminants into drinking water sources, the speed of contamination detection, and water suppliers' and consumers' reactions. A Monte Carlo analysis is conducted to address the uncertainty of the parameters in the event tree and related to the illness and averting costs. We find that significant economic costs arise due to climate change and that society may be willing to pay a large sum to ensure uncontaminated water supplies. The results can be used to aid in choosing optimal adaptation and mitigation strategies in the face of climate change.

PHA066 Environmental Degradation of Gobeid Habitat in Grenada. Dorfman, D.*, Monmouth University, West Long Branch, NJ. *Sycidium palmieri* is a gobeid fish that lives in the fresh and saltwater of the Caribbean. A study of this species was made in Grenada from anecdotal data, literature, field collections, and visits to fish markets over a six month period during 1997-1998. This fish is catadromous. That is, it spends most of its life in rapidly flowing freshwaters and then migrates to the sea, between October and January, to spawn. The larvae remain in saltwater for a short time after hatching, then migrate to freshwater. As these fishes, now in their postlarval stage, enter freshwater rivers and streams they are collected in large numbers. The fishes lack pigment and are referred to as titiree. These are rinsed, then mixed with rice and formed into fish cakes. This delicacy is then cooked and eaten. Surviving fish develop pigment in a few days and they migrate up rapidly flowing water by darting from boulder to boulder and maintaining their hold by a sucker on their ventral surface, formed by their pelvic fins. These streams are polluted primarily by soap, and by occasional industrial discharges of untreated wastes. The human population is growing at a low rate (0.72%). However, plans for increased building of hotels to attract greater numbers of tourists may destroy fish habitat, including that of *S. palmieri.* Subsidizing the use of biodegradable soaps, laws to control effluent discharge, and secondary treatment of sewage may maintain fish habitat.

PHA067 Ultraviolet Induced Phototransformations of Dissolved and Colloidal Organic Matter. Tarr, M.A.*, Wang, W., University of New Orleans, New Orleans, LA; Bianchi, T.S., Engelhaupt, E., Tulane University, New Orleans, LA. Several important properties of dissolved and colloidal organic mater (DOM and COM) may be strongly influenced by photoprocessing in the environment. Previous studies have illustrated the photoproduction of low molecular weight carbon species from DOM, and more recent work has indicated ammonium and amino acids as photoproducts. The release of these photofragments can have a significant impact on phytoplankton and zooplankton populations. Ultimately, these impacts can potentially affect entire ecosystems as well as global climate. In addition to release of small photoproducts, the photoprocessed DOM/COM molecules are structurally altered. DOM/COM solubility, light absorption properties, photosensitization, metal binding, and chemical reactivity may all be altered by phototprocessing. In order to better understand the impacts of these changes, we have studied the dynamics of nitrogen photoproduct formation as well as phototransformations of COM structure as followed by ^{13}C NMR. For several DOM/COM samples, we have observed zero order ammonium photoproduction with higher rates at low pH. Formation of amino acid products has also been observed. ^{13}C NMR data form COM has indicated a decrease in aromatic and carbonyl functional groups after photoprocessing.

PHA068 Photochemical reactivity of dissolved lignin in river and ocean waters. Opsahl, S.*, U.S. EPA, Athens, GA; Benner, R., University of Texas at Austin, Port Aransas, TX. About 0.25 * 10^{15} g of terrigenous dissolved organic matter (DOM) is discharged by rivers to the oceans annually. Recent studies using dissolved lignin as a molecular biomarker reveal that most riverine DOM is oxidized on time scales of months to years. Photochemical and microbial degradation are both mechanisms which contribute to the remineralization of terrigenous DOM and the relative importance of these processes was examined in a series of light/dark bottle experiments. DOM in river water was highly bleached after 28 days of exposure to sunlight. The dissolved lignin component was found to be photoreactive and decreased substantially in concentration. Dissolved lignin also underwent a major shift from high molecular weight (HMW; >1,000 Dalton) to low molecular weight (LMW; <1,000 Dalton) DOM as a result of photochemical processes. This provides direct evidence that specific DOM components can undergo dramatic reductions in size yet retain their molecular identity. Notably, the fraction of dissolved lignin in river water which remained at the end of the experiment appeared to resist further photochemical oxidation. HMW DOM from open-ocean seawater did not undergo photobleaching, and the dissolved lignin component resisted photochemical oxidation. Furthermore, the molecular composition of open-ocean dissolved lignin was similar to the photochemically-altered riverine material. These findings suggest that photochemical reactions play a prominent role in determining the composition, reactivity and fate of terrigenous DOM.

PHA069 Interactive Photochemical and Microbial Degradation of Dissolved Organic Matter. Zepp, R.*, US EPA, Athens GA; Moran M. and Sheldon, W., U. Georgia, Athens, GA. Colored dissolved organic matter (CDOM) controls the penetration of solar UV radiation in freshwater and marine environments and interferes with the remote sensing of color in coastal regions. We have observed large variations over time and space in the concentrations, bacterial degradation rates and the UV-visible absorption spectra of CDOM in southeastern U.S. estuaries. Variation in bacterial degradation rates of up to 3-fold were measured among adjacent estuaries; similarly, variations of up to 10-fold were measured temporally within a single estuary. Moreover, slopes of log plots of CDOM absorbance versus wavelength increased significantly with increasing salinity in the estuaries. Both photooxidation and photochemically-mediated microbial degradation of the CDOM contribute to these variations in spectral and biological properties. Photooxidation of the CDOM results in loss of absorbance in the UV and visible spectral regions and altered CDOM that is more susceptible to microbial degradation. The variability also is attributable in part to iron catalysis of CDOM photooxidation, a process that is most important at the freshwater end of the estuaries. Moreover, photooxidation leads to the coagulation of the iron and CDOM, a process that has significant effects on the absorption and scattering properties of irradiated estuarine waters.

PHA070 Photochemical Degradation of DOM in Two Nova Scotian Lakes. Moore, R.J.*, Dalhousie University, Halifax, Nova Scotia, Canada; Miller, W.L., Dalhousie University, Halifax, Nova Scotia, Canada. Water samples from Beaverskin and Pebbleloggitch Lakes were collected from June 1997 through September 1997. These lakes experience the same microclimate yet exhibit radically different light absorption properties. Samples were analyzed for absorptivity to evaluate colour loss over the course of the summer due to photooxidation of CDOM. They were also evaluated for photochemical production rate of CO_2 in laboratory irradiations of 0.2 µm filtered water samples using full spectrum simulated sunlight. Measurement of CO_2 production during the irradiation period employed a stripping system and a Licor infrared CO_2 analyzer. Wavelength-dependent quantum yields for CO_2 production were obtained by examining production differences between samples exposed using sequential long-pass cut-off filters and a xenon-lamp irradiation system. During the drought conditions of 1997, absorptivity values decreased by 50% in the highly coloured Pebbleloggitch Lake and by 80% in the clear Beaverskin Lake. Absorbance normalized photochemical production rates of CO_2 were found to be greater than CO production rates published for similar lakes. Quantum yield spectra exhibit a similar shape to CO spectra found in the literature. Photooxidation of CDOM in lake waters does lead to significant fading and CO_2 production is a significant pathway for photochemical breakdown of CDOM.

PHA071 Modelling Environmental Photochemical Processes Using SolarFx. Valentine, R. L.; Thornberry, T., Department of Civil and Environmental Engineering, University of Iowa, Iowa, City, Iowa We have developed a comprehensive model of use in predicting solar influences on several important aquatic photochemical processes. These include photochemical oxidation of anthropogenic compounds and natural organic matter, and the photochemical fading occurring in water as a result of the loss of chromophores in humic matter. One unique aspect of this model is that it has a time resolution of approximately one minute which makes it useful in conducting real-time experiments utilizing exposure to natural sunlight. The solar irradiance submodel accounts for changes in solar light flux as a function of global position, elevation, ozone concentration, water vapor, and particulate matter. It is used to drive several aquatic process submodels. The theoretical basis for the model will be presented and model calibration and validation results will be shown which utilize natural sunlight for: 1) transformation of a standard photochemical actinometer, 2) formation of carbon monoxide from photochemical oxidation of humic substances, and 3) photochemical fading of natural water. The model will be used to explore factors influencing and limiting photochemical transformations in general such as light attenuation, depth of water column, and the influence of mixing.

PHA072 Application of a Chemiluminescence Method for the Analysis of H_2O_2 in Surface Waters with Varying Natural Organic Matter Concentrations. Moegling, J. K.*, Cooper, W. J. and Kiddle, J. J. University of North Carolina at Wilmington, Wilmington, NC 28403-3297. Hydrogen peroxide, H_2O_2, is a relatively stable product of natural humic substances photolysis in surface waters. There are numerous methods for the determination of sub-micromolar concentrations of H_2O_2 in natural waters. One of the most popular is the fluorescent decay method using scopoletin. However, a major drawback to this method in natural waters is that the fluorescent signal from the scopoletin overlaps with that of naturally occurring organic humic substances. Fan and Huisheng (*Fenxi Huaxue*, 1992, 20(3) 342-344) reported an alternate chemiluminescent method for determining trace amounts of H_2O_2 in water using an acridinium ester (10-methyl-9-(p-formylphenyl)-acridinium carboxylate fluorosulphonate). This compound is not commercially available. And, no complete synthesis of the compound has been published. We have successfully synthesized the ester and found it to react as reported above. The first portion of this paper will briefly outline the synthetic route to the product. The second portion of this paper will describe our initial studies on the application of this chemiluminescent method for the determination of H_2O_2 in surface waters. The chemiluminescence reaction requires no catalyst or metal ion complexes. It is a simple and rapid reaction that is selective, sensitive and linear over a wide range, and, suitable for use in field analyses. This paper describes this method and the results we have obtained in several different environments with varying concentrations of naturally occurring humic substances. The environments that have been studied include relatively highly colored river water, near shore and coastal waters, open ocean areas and rain water. We have found the distribution of H_2O_2 is similar to those that have been published. We feel it will be possible to extend this method to the on-line, continuous determination of H_2O_2 in natural waters with varying organic matter concentrations.

PHA073 Gradients of DOM and optical properties in Lake Superior. Green, S.A., Michigan Technological University, Houghton, MI. Lake Superior is a highly transparent lake with a[350nm]<1.5 /m). However, many of its tributaries are highly colored (a[350nm]=10 to 50 /m). Dissolved organic matter (DOM) shows similar gradients from rivers (6-20 ppm) to the central lake basin (1-2 ppm). The observed gradients and DOM distributions are a result of coupling of physical, chemical, and biological processes in the nearshore zone. The relative importance of each mechanism will be analyzed on both local and basin-wide scales.

PHA074 Acid-induced Changes in DOC Quality and Optical Environment in an Experimental Whole-lake Manipulation. Donahue, W.F.* & Schindler, D.W., University of Alberta, Edmonton, AB, Canada; Page, S.J. & Stainton, M.P., Freshwater Institute, Winnipeg, MB, Canada. Fluorescence analyses of water samples, archived from 1985 to 1997, were used to typify DOC quality in experimentally acidified lakes and reference lakes at the Experimental Lakes Area, in northwest Ontario. Carbon-specific DOC fluorescence (CSF) during peak acidification (pH 4.5) was 40-50% of that of reference lakes. Reference lakes showed similar, but smaller changes in CSF during several years of prolonged drought in the late 1980s. This is consistent with changes in fluorescence-peak geometry, which indicate a switch in quality of DOC during acidification. The acid-induced change in DOC quality was likely due to increased chemical oxidation of the UV-absorbent aromatic portions of allochthonous DOC molecules, leaving more UV-transparent aliphatic chains. UV-absorbance properties of DOC during peak acidification resembled those of alpine lakes rather than boreal lakes. In addition to causing decreased [DOC], the change in the nature of DOC from allochthonous to "autochthonous-like" following acidification and drought may have an important role in physical, biological, and chemical processes within these lakes. With recovery from acidification, DOC quality has also recovered to that representative of boreal shield lakes.

PHA075 The Photobleaching of Colored Dissolved Organic Matter in Natural Waters. Del Vecchio, R.*, Vodacek, A. and Blough, N.V., Department of Chemistry and Biochemistry, University of Maryland, College Park, MD 20742, USA. The effects of monochromatic UV and visible radiation on the optical absorption and emission properties of colored dissolved organic matter (CDOM) were examined for a Suwanee River Fulvic Acid (SRFA) standard and for water from the Delaware and Chesapeake Bays. The primary bleaching occurs at the irradiation wavelength for both the absorption and the fluorescence. Less efficient bleaching occurred outside the irradiation wavelength. Fluorescence always decreased more rapidly than absorption during the first hours of irradiation, leading to a rapid initial decrease in fluorescence quantum yield. Thereafter the fluorescence and absorption decay similarly and the fluorescence quantum remains relatively constant. In all cases the loss of absorption and fluorescence with time were fit well as a sum of two exponential decays. The rate constants for the absorption loss at the irradiation wavelengths (296, 313, 334, 366 and 407nanometer) were in the range between $4x10^{-5}$ h^{1} and $7x10^{3}$ h^{1} for the slow decay and in the range between $4x10^{2}$ h^{1} and $6x10^{1}$ h^{1} for the fast decay. The intensity of the Xe-Hg lamp at those wavelengths varied from $5x10^{-4}$ to $4x10^{-3}$ Watts per centimeter^{-2}.

PHA076 Photochemical formation of NMHCs (non-methane hydrocarbons) from marine dissolved organic matter: can we write a mechanism? Milne, P.J.*, RSMAS, University of Miami, Miami, FL., Riemer, D.D., NCAR, Boulder, CO, Pos, W.H., University of Sao Paolo, Brazil, and Zika, R.G., RSMAS, University of Miami, FL. Direct photoproduction of a range of non-methane hydrocarbons, especially low molecular weight alkenes, has long been demonstrated in seawater [Wilson et al., 1970]. Although their mixing ratios in remote marine atmospheres are small (sub ppbv or less) these compounds are highly reactive and may be important photochemical ozone precursor compounds. Due to our incomplete knowledge of the corresponding magnitude of the sea-air fluxes, the net impact of these compounds on the oxidizing capacity of the marine atmosphere and their importance in global carbon cycling remains uncertain. Recent field and laboratory investigations of the photochemical formation of alkenes, alkanes, and isoprene including their photobiologically mediated production in sunlit surface waters provide a number of useful insights into the several parameters that control their production. Some of these factors to be discussed include the wavelength and substrate concentration, the irradiation conditions and sample treatment, such as filtration, the presence or absence of oxygen. Suggested mechanistic pathways to the formation of compounds via carbonyl mediated Norrish Type II reactions helps deepen our understanding of NMHC photoproduction mechanisms and may, through a consideration of the observed product ratios, provide some information as to the structure of marine DOM.

PHA077 Photochemical Formation of Hydroxyl Radical by Colored Dissolved Organic Matter and Substituted Quinones. Vaughan, P. P.*; Blough, N. V., University of Maryland, College Park, MD. Previously, the wavelength dependence of the quantum yield for photochemical hydroxyl radical (OH) production was determined for Suwannee River fulvic acid samples, employing a nitroxide probe to trap the methyl radical produced by reaction of OH with dimethyl sulfoxide. Catalase experiments show that Fenton chemistry can account for at most 50% of the total signal under aerobic conditions for SRFA irradiated at 310 and 320 nm. These results indicate the presence of a dioxygen-independent pathway of hydroxyl radical production that cannot be assigned to nitrate/nitrite photolysis or to Fenton chemistry. Furthermore, it has been proposed that quinone moieties present in CDOM may be responsible for photochemical OH formation. Evidence suggests that OH forms from a photochemically generated, excited triplet state of benzoquinone that undergoes hydrogen atom abstraction from water. Using nitroxide trapping, the quantum yield for the formation of OH, at 424 nm, from p-benzoquinone has been determined and is in good agreement with the value reported by Ononye and Bolton for formation of BQ.-. Quantum yields for the formation of OH by a number of substituted quinones are reported and interpreted.

PHA078 Use of Quadricyclane as a Probe for Superoxide. Hassett, J.P.*, Beretvas, M.K. State University of New York – College of Environmental Science and Forestry, Syracuse, NY 13210. Burns, S.E. University of Western Michigan, Kalamazoo, MI 49008. Quadricyclane, a hydrocarbon with highly strained cyclopropane and cyclobutane rings, undergoes indirect photolysis in natural waters. Experiments suggest that superoxide / hydroperoxy radicals produced by DOM are reacting with quadricyclane. The wavelength dependent production of superoxide by DOM was followed by measuring the degradation rate of quadricyclane at four wavelengths (using narrow bandpass filters) and factoring out the fraction of light absorbed by the humic material. The efficiency of production decreases by a factor of five over the range 313 – 435 nm. Experiments using cutoff filters have shown that degradation of quadricyclane takes place to a significant degree in the visible region. The use of superoxide probes with known rate constants allowed an apparent quantum yield for superoxide production to be calculated and this value was compared for different waters. Initial results have demonstrated that quadricyclane exhibits slower reaction rates in natural waters than in an Aldrich humic acid solution.

PHA079 The Hydrated Electron as a Possible Sink of Halogenated Organic Compounds in Natural Waters. Thomas-Smith, T. E., Blough, N.V., University of Maryland College Park, College Park MD. There is some evidence to suggest that the photoproduction of the hydrated electron (e^- (aq)) by colored dissolved organic matter CDOM could act as a sink for halogenated hydrophobic organic compounds via heterogeneous reductive dehalogenation. The wavelength dependence of the quantum yields for e^- (aq) formation was determined by a new method using nitroxide trapping. The wavelength dependence of the quantum yield for e^- (aq) production from SRFA shows a decrease in quantum yield with increasing wavelength with values ranging from $1x10^{-5}$ at 366nm to $2x10^{-4}$ at 296nm. Recent work on the reductive dehalogenation of both soluble and hydrophobic halogenated compounds will also be reported.

PHA080 Humic Substances in Biotransformation of Xenobiotica. Klavins, M., Serzane,J. Department of Environmental Sciences, University of Latvia, Raina blvd. 19, LV 1568, Riga, Latvia. Humic substances (HS) much influence soil fertility, and they play a principal role in the turnover of organic carbon. The interaction of humic substances with xenobiotics may modify the uptake and toxicity of these compounds by aquatic organisms, and affect the fate of pollutants in the environment. Of the humic properties which have been little analyzed is their catalytic activity, though HS contain many groups and structures which in similar macromolecules are responsible for their catalytic activity. In the present study the catalytic activity of humic substances of different origin in the reactions of hydrolysis of esters and condensation of carbonylcompounds has been studied. These reactions can be of importance with regard to the fate of environmental pollutants. Humic substances demonstrate appreciable impact on the fate of organic xenobiotics in natural environments. Of the humic substances, the aquatic fulvic acids are the most active in hydrolysis reactions, but the velocity of studied reaction depends also on temperature, the concentration and of humic substances used, as well as presence of surfactants thus indicating possible micellar catalysis presence.

PHA081 Use of Water Table Management Systems for Microbial Bioaugmentation of Contaminated Soils. Mehmannavaz, R.*, Prasher, S.O., Macdonald Campus of McGill University, Ste-Anne de Bellevue, Quebec, Canada; Ahmad, D., INRS-Santé, Universite du Quebec, Pointe Claire, Canada. Microbial bioaugmentation of some contaminated sites, such as those with PCBs seems to be becoming more important. PCBs require different groups of organisms for their complete degradation and it is possible that the indigenous microorganisms do not have the ability to degrade such chemicals efficiently. Therefore, bioaugmentation could serve as an alternative to other available technologies for bioremediation purposes. However, a major problem for in situ bioaugmentation of contaminated soils is that the delivery of desired microorganism to deeper zones of a contaminated site has not met with much success. In this study, bioaugmentation of pristine and PCB contaminated sandy loam soils was conducted in large stainless steel columns, 1000 mm in length x 200 mm in diameter. Delivery and implantation of bacteria were observed in both soils using a Water Table Management (WTM) system. The system allowed a uniform distribution of bacterial cells throughout the soil profile, from the surface up to depths of 850 mm. It was observed that under saturated conditions, bacterial cells could be transported by the head of the water flow to different depths throughout the soil profile and be distributed. Furthermore, the WTM system proved to be an effective method for creating aerobic and anaerobic conditions in the soil profile. Our results indicated that WTM system is a promising technology for bioaugmentation in in situ soil decontamination tasks.

PHA082 Diversity Pattern of Microbial Populations in a PCB Contaminated Soil During a Bioremediation Treatment. Mehmannavaz, R.*, Prasher, S.O., Macdonald Campus of McGill University, Ste-Anne de Bellevue, Quebec, Canada; Ahmad, D., INRS-Santé, Universite du Quebec, Pointe Claire, Canada. During bioremediation of contaminated soils, determining the number and type of indigenous microorganisms plays an important role. It is helpful to see what and how some microorganisms can tolerate and adapt to changes in the ecological factors that might occur during a bioremediation process. In this study, we attempted to establish the microbial population diversity in a soil contaminated with mixtures of PCBs (Aroclor 1242, 1248, 1254, and 1260), diesel, and heavy metals during a bioremediation process. This soil was bioaugmented with an isolated strain, Comomonas, and an Indigenous mix culture, as separate treatments. These treatments were exposed to periodical aerobic and anaerobic conditions by saturating and draining of the soils for a period of 344 days. The initial and final indigenous microbial populations were determined on 4 different nutrient media, TYc, TYc/100, TYct, and MH. The TYct nutrient plates showed the least diversity in the microbial populations in different treatments with a maximum of 4 different colony types in the treatment augmented with Comomonas. However, the MH nutrient plates showed up to 5 different colony types in the treatments augmented with the isolated strain, and the Indigenous mix culture. Furthermore, MH plates showed up to 9 different colony types in total with all the treatments, whereas, TYc and TYc/100 showed 7 different colony types each for the period of the treatments. The results indicated that the indigenous microbial populations were affected by the different bacterial cultures used to bioaugment the soil and the population size and diversity varied during the bioremediation process.

PHA083 Microbial Degradation of Oil Hydrocarbons in Lake Baikal. Dagurova, O.P., Kozyreva, L.P., Barkhutova, D.D., Namsaraev, B.B*.Laboratory of Microbiology, Institute of General and Experimental Biology of SB RAS, Ulan-Ude, Russia. The oil pollution of water reservoirs disturbs the biological equilibrium as it gives toxic influencing on the biota. The natural hydrocarbon-utilizing bacteriocenosis plays the leading role in the processes of self-cleaning. In present investigation the quantities of hydrocarbon-oxidazing microorganisms were determinated in water, shallow-water sediments and deep-water sediments. The quantity of in surface water reaches 10 thousands cells/ml, where as it make up 10 cells/ml in deep water and shallow water. The number 1-10 thousands cells/ml was founded in shallow sediments that is higher on an order than one was found in deep-water sediments (from 10 to 1000 cells/ml). Maximum value - 100 thousands cells/ml observed near Severobaikalsk town can be comparable with cells' numbers of oil-polluted water and sediments of the Selenga River. The enrichment and pure cultures were obtained from shallow sediments. Some of them are able to grow due to different aliphatic and aromathic substrates. They have shown a rapid growth at high concentration of substrates in vitro (10%) and low temperature (4 C). These rezults show that hydrocarbon-utilizing bacteria degrade natural and anthropogenic oil pollution in water, shallow sediments and deep bottom sediments in the Baikal.

PHA084 Microbial Transformation Products of Atrazine in Estuarine Sediments. Patel, P. P.*and Aelion, C. M., USC-Columbia, Columbia, SC. Atrazine is a triazine herbicide that has been widely used in the U.S. for decades to control broadleaf and grassy weeds on fallow and industrial lands and in agricultural production. Atrazine does not readily sorb onto soil particles, however, its metabolites may have a higher affinity for sediment than for water and so it is crucial to know the presence of any such polar and non-polar metabolites. Also, atrazine has a long half-life that allows it more time to undergo biotransformation. The main objective of this study was to determine the microbial production of polar and non-polar metabolites of atrazine in sediment samples. Three sites near Georgetown, SC with varying degrees of land uses were chosen for this study–an undeveloped forested area, a suburbanized area, and a country club that receives large inputs of pesticides and fertilizers. Laboratory experiments conducted using sediment microcosms found minimal microbial mineralization or cellular incorporation of ^{14}C atrazine by measuring $^{14}CO_2$ production in the incubation vials using a Liquid Scintillation Counter. Mineralization of ^{14}C atrazine was significantly different among the sampled sites depending on the organic content and exposure of the sediments to atrazine. Solid Phase Extractions were performed to collect polar, to moderately polar, to non-polar compounds using solvents of decreasing polarity. Degradation products of atrazine are of interest due to their potential phytotoxicity. The identity of these extracted compounds is currently under investigation. Preliminary results indicated that by day 54 of incubation, ^{14}C radioactivity significantly decreased in the liquid portion and in methanol extracts of sediments, and the portion irreversibly sorbed to sediments increased. Atrazine seems to be resistant to mineralization but may be biodegrades in South Carolina's estuarine sediments.

PHA085 Using Respiration Rates and Stable Carbon Isotopes to Monitor the Biodegradation of Orimulsion by Marine Benthic Bacteria. Lapham, L. L., Chanton, J. P.*, and Proctor, L. M., Florida State University, Tallahassee, FL. Respiration rates and $^{13}CO_2$ ratios of respired CO_2 were measured in marine sediment slurries to evaluate the biodegradation of Orimulsion, a fuel consisting of 70% Cerro Negro bitumen, 30% water and 0.1% surfactant. Three experiments contained Orimulsion while two contained bitumen alone. All five experiments contained marine sediment as both a bacterial inoculum and a carbon source and assayed over 21 days. Isotopic signatures of marine organic matter (MOM) and hydrocarbons differed: MOM, -23.84 ± 0.56‰; Orimulsion, -27.46±0.12‰; bitumen, -27.27±0.11‰; and surfactant, -27.94±0.10‰~. We hypothesized that respiration rates would be higher and that the isotopic signature of the respired CO_2 would be ^{13}C depleted in Orimulsion/bitumen slurries relative to controls. Over 21 days, hydrocarbon degradation was apparent. Bacterial numbers increased but stabilized to 10^9 cells/g dry weight sediment within 7 days. However, respiration rates in the hydrocarbon-amended slurries continued to increase to 1.4-1.8 times above controls. Since the greater surface area of the hydrocarbon slurries alone could enhance respiration, we measured the $\delta\,^{13}C$ of the CO_2 to directly demonstrate hydrocarbon degradation. The isotopic ratio of the CO_2 in the Orimulsion-amended slurries was 2 to 3‰ ^{13}C depleted relative to controls. Although lower, the isotopic ratio in the bitumen amended slurries was 0.1 to 0.5‰ ^{13}C depleted relative to controls. From mass balance estimates, we determined that the isotopic ratio of the CO_2 respired due to MOM was -20.0 to -22.8‰ whereas additional CO_2 respired in the hydrocarbon-amended slurries was -25 to -27‰, confirming hydrocarbon degradation.

PHA086 Enhanced Co-Metabolism of Cerro Negro Bitumen with Amendments of Natural Marine Carbon Substrates. Cherrier, J., FAMU, Tallahassee, FL, Lapham, L. L., Chanton, J. P., and Proctor, L. M., Florida State University, Tallahassee, FL. Aerobic respiration and $^{13}CO_2$ ratios of respired CO_2 were measured in sediment-free preparations of benthic marine bacteria to evaluate biodegradation of Cerro Negro bitumen. All bottles contained bacteria extracted from marine sediments as an inoculum and were incubated 21 days. Amendments were made with natural marine carbon substrates of differing isotopic signature: marine sediment organic matter, MOM, (-23.84 ± 0.56‰), seagrass (-10.7‰), or pinfish (-15.8‰). Experimental bottles contained the hydrocarbon, bitumen (-27.27 ± 0.11‰). We hypothesized that respiration and co-metabolism of bitumen would increase with the addition of labile carbon substrates and that the isotopic signature of the respired CO_2 would be ^{13}C depleted in bitumen-containing bottles. At the end of 21 days, the amount of CO_2 respired from bitumen only with no amendments was 1600 ppm CO_2; MOM amendments elevated bitumen degradation by 500 ppm CO_2; seagrass amendments elevated bitumen degradation by 1400 ppm CO_2; and pinfish amendments further elevated bitumen degradation by 18400 ppm CO_2. The isotopic ratio of the respired CO_2 due to either the bitumen with no amendments or bitumen with MOM amendments was 4‰ ^{13}C depleted relative to the bitumen-free controls, bitumen with seagrass amendments were 7‰ ^{13}C depleted relative to the seagrass only controls, and bitumen with pinfish amendments were 3‰ ^{13}C depleted relative to the pinfish only controls. Over 21 days, bitumen biodegradation was enhanced by the addition of these native marine carbon substrates and this increased biodegradation correlated with the increasing lability of the carbon substrates.

PHA087 Biodegradation Rates of Diesel Hydrocarbons. Olson, J.J.*, Mills, G.L., Herbert, B.E., and Morris, P.J., Savannah River Ecology Laboratory, Aiken, SC. Studies with complex mixtures, like whole fuels, have shown that hydrocarbons in petroleum do not follow a simple degradation sequence and the biodegradation rates of specific compounds vary among fuels. These studies indicate that the interactions between hydrocarbons of different classes influence the rates of biodegradation of specific compounds as well as entire classes. However, without data on the rates of biodegradation of the compounds and classes independent from the complex mixture it is difficult to extrapolate this data to use as a predictive tool for other fuels or as fuel-composition changes due to weathering. The biodegradation of specific compound classes isolated from complex mixtures versus composite hydrocarbon mixtures have not been reported. Three hydrocarbon classes (n-alkanes, branched & cyclic alkanes, and aromatics) were separated from a diesel fuel. The biodegradation rates of each class and specific compounds within each class were quantified over a 35 day period in microcosms that contained one of the three classes and in micorcosms that contained an assemblage of the three classes that resembled the original diesel fuel (composite solution). Triplicate microcosms were sampled on experimental days 0, 3, 7, 15, and 35. Microbial populations were quantified by total hetrotrophic plate counts. Hydrocarbon concentrations were quantified by GC/FID and specific compound identification were confirmed by GC/MS. After 35 days, the relative biodegradation for each hydrocarbon class was *n*-alkanes> aliphatic fraction of composite solution> aromatics> branched & cyclic alkanes> aromatic fraction of composite solution. The biodegradation rates ranged from 200 µg/day to 4,500 µg/day.

PHA088 Bioavailability and Bioremediation of Co-occurring PCBs and Mercury in Saltmarsh Sediments. Maruya, K.A.*, Lee, R.F., Frischer, M.E., Kostka, J.E., Windom, H.L. Skidaway Institute of Oceanography, Savannah, GA. Saltmarsh sediments at a Superfund site in coastal Georgia (USA) have been contaminated with PCBs (as Aroclor 1268) and mercury for several decades, causing periodic fish and shellfish harvesting closures. Our multidisciplinary team has sought to understand the governing processes that control the bioavailability and in situ transformation of PCBs and Hg. Our ultimate goal is to develop in situ remediation strategies that better preserve the natural saltmarsh environment. GC-ECD and GC-MS analyses of sediment cores indicate that reductive dechlorination is occurring in the *Spartina* rhizosphere, albeit at a very slow rate. Both contaminants accumulate in marsh biota and may act in concert to cause DNA damage and embryotoxicity in grass shrimp. Biogeochemical measurements in the rhizosphere indicate that sulfate reduction is the dominant pathway for carbon mineralization. 16S rRNA probes designed for sulfate reducing bacteria (SRB) suggest that different groups may predominate in different horizons of the rhizosphere. Mercury methylation rates correlated with sulfate reduction rates and also with the abundance of specific groups of SRB. On a pilot scale, we plan to test PCB and Hg biotransformation strategies that enhance the activity and synergy of the existing microfloral assemblage using our Bioremediation and Environmental Mesocosm (BERM) facility, a series of custom-built, state-of-the-art flow-through analog tanks.

PHA089 Relationship Between Carbofuran-Degrading Microbial Populations and Carbofuran Degradation in Enhanced Soil. Trabue, S.L.*, DuPont, Wilmington, DE; Orgam, A.V., University of Florida, Gainesville, FL; and Ou, Li-Tse, University of Florida, Gainesville, FL. After two consecutive annual applications of carbofuran, the degradation rate of the chemical was enhanced in surface and subsurface soils from a site in Florida. Carbonyl-labeled (CAL) ^{14}C-carbofuran was used for determining the hydrolysis rate in soil, and uniformly ring labeled (URL) ^{14}C-carbofuran was used for determining the mineralization rate and degradation pathway in soil. The population sizes of carbofuran-degrading microorganisms were enumerated using a ^{14}C-MPN (most-probable-number) technique. The hydrolysis rate of carbofuran in the enhanced soil increased with an increase in the number of annual applications of the chemical. At the same time, the population size of carbofuran hydrolyzers in soil also increased. However, neither the mineralization rate of URL-^{14}C-carbofuran in enhanced soil nor the population size of carbofuran ring degrading microorganisms increased following four annual applications of the chemical. Although URL-^{14}C-carbofuran in enhanced soil was rapidly mineralized, the population size that mineralized URL-^{14}C-carbofuran was very small. Due to the lack of growth in the carbofuran ring degrading population, the ring structure of carbofuran in the enhanced soil appeared to be degraded cometabolically. The population size of carbofuran-hydrolyzing microorganisms was significantly larger than the population size that mineralized the ring structure of carbofuran. Carbofuran in enhanced soil was degraded mainly through hydrolysis, likely without undergoing oxidation to 3-hydroxycarbofuran and 3-ketocarbofuran.

PHA090 Biotransformation of the Polycyclic Musk, HHCB, during Sewage Treatment. Itrich, N.R.*, Simonich, S.L. and T. W. Federle. The Procter and Gamble Company, Cincinnati, Ohio. Polycyclic musks (PCMs) are important fragrance ingredients, which are used at low concentrations in a variety of consumer products. One of the most commonly used PCMs is HHCB (1,3,4,6,7,8-hexahydro-4,6,6,7,8,8-hexamethyl-cyclopenta[γ]2-benzopyran). To better understand HHCB fate in the environment, biotransformation was examined under realistic conditions in activated sludge. A novel approach that involved tandem filtration and solid phase extraction was developed to efficiently recover parent and metabolites from mixed liquor. In addition, the test system was designed to measure volatilization of parent and evolution of 14CO2. 14C-HHCB was dosed at a low level (50 ug/L) to freshly collected activated sludge, and disappearance of parent and formation of metabolites were monitored over time. In the abiotic control, recovery of HHCB was initially > 90% but decreased over time with the appearance of a polar metabolite (#1) that co-eluted with a lactone standard. Losses due to volatilization were low by comparison. In the bioactive treatment, parent HHCB decreased more quickly concurrent with appearance of the presumptive lactone and subsequent appearance of a more polar materials (#2) that co-eluted with a hydroxy acid standard. No significant volatilization was observed in this treatment, and the half-life for HHCB parent was estimated to be approximately 35 hours. Rad-HPLC was used to estimate the Kow of the parent and metabolites in the activated sludge extracts. Measured Kow values were 5.9 for HHCB, 2.3-3.0 for metabolite 1, and ~0 for metabolite 2. This work demonstrates that HHCB is biotransformed during wastewater treatment to polar metabolites, that have substantially lower Kow values than the parent molecule. Thus, biotransformation and abiotic oxidation processes have the potential to significantly reduce the toxicity and bioconcentration of HHCB in aquatic environments.

PHA091 Hydrogen Sulfide Biotransformation in Hot Springs of Baikal Region. Smirnyagina S., Danilova E., Namsaraev B.* Buryat Institute of General and Experimental Biology, Ulan-Ude, Russia. Hydrogen sulfide is known to be present at hot alkaline springs waters and microbial mats in so- luble state or as HS^- and S^- anions. It has a toxic effect for living organisms. In this study we investigated microbial transformation of hydrogen sulfide in samples of hot springs. Sampling sites were loca- ted in the South-west Baikal shore zone and in the Barguzin Basin of Eastern Baikal sea-line zone. Most thermal fluids in warm slightly al- kaline (pH 7) springs located in the South-west were enriched in car- bon dioxide and nitrogen. Sulfide concentrations were not over 2 mμ. Temperatures averaged 30 °C. More hyperthermal silicious nitrogenous alkaline (pH 8.5-9.9) springs where temperatures averaged 50, 60 and 70 °C occured in the Barguzin Basin. Most hydrothermal fluids were en- riched in nitrogen, oxygen and hydrogen sulfide. The highest concent- rations of sulfide (31-35 mμ) occured in Umhey and Kucheger Springs. Sulfide in hot springs can be produced both abiotically and microbial- ly. Possible abiotic source of sulfide is injection of volcanic gases. An alternative source could be biological production by principally sulfate-reducing bacteria. Cell densities of sulfate-reducter Desulfo- bacter sp. in hot spring samples cultivated in Viddel medium were fo- und to be 100-1000 cells/ml. The main sulfide consumer in hydrothermal microbial communities were presented by photosynthesysing anaerobic bacteria, sulfur- and thiobacteria. The latter oxidizes hydrogen sul- fide to elementar sulfur. In sulfur microbial mats dominating forms were found to belong to the genera Thiothrix, Thioploca, Phormidium. Thiobacteria cell densities in the Bejerink medium were 1000-10000 cells/ml.

PHA092 HPLC Methods for Biodegradation Studies of the Pharmaceutical, BRL 49653-C. Vaughan, P.B., Ziegenfuss, P.S., Brum, J.L., Hannah, R.E., SmithKline Beecham, King of Prussia, PA. Biodegradation studies present unique problems for liquid chromatography methods development. We determined the rate of aerobic biodegradation of the pharmaceutical, BRL-49653-C, in an activated sludge matrix. Previous attempts to analyze for BRL-49653-C in this matrix provided poor results due to retention-time shifts of the analyte peak. It appeared that extra-cellular microbial secretions interacted with the solid phase in the analytical column, changing it's retention characteristics. A study was undertaken to devise a method that offered consistent results with a fixed retention time and satisfactory resolution of the analyte peak. Aqueous portions of activated sludge spiked with BRL 49653-C and an unspiked control were sampled using Anotop® (0.2μm) filters. Samples were quantified using liquid chromatography. Alterations to the original method included; a change in analytical column, decreased ionic strength and increased organic modifier concentration of the mobile phase, and changing from a gradient to an isocratic run. The use of filtering devices coupled with these modifications resulted in an analytical method which provided consistent analyte retention times and a total run time of only 15 minutes. Possible rationale include enhanced filtration of interferences and a higher organic modifier concentration, which decreased analyte interactions with the solid phase. Conclusions provided by the modified analytical method showed that the disappearance of BRL 49653-C approximated first-order kinetics, with an observed half-life of 1.2 days. One biodegradant was observed by HPLC and identified by LC/MS as a des methyl analog.

PHA093 Assessing the Environmental Persistence of Bisphenol A. West, R.J.*; Goodwin, P.A., Klecka, G.M., The Dow Chemical Company, Midland, MI.; Dorn, P.D., Battersby, N.S., Shell Development Company, Houston, Texas; Staples, C.A. Assessment Technologies Inc., Fairfax, VA. The biodegradability of bisphenol A was measured in various tests to determine the fate and lifetime of the compound during wastewater treatment and in surface water environments. As a stringent means of determining the potential fate of bisphenol A during wastewater treatment and in natural environments, the ready biodegradability of the compound was evaluated using the Organization for Economic Cooperation and Development (OECD) Method 301F: Manometric Respirometry Test. Biodegradation of bisphenol A resulted in 81 to 93% of theoretical oxygen demand (ThOD) consumed after 28 days, and >77% ThOD consumed within 10 days of biodegradation onset. Extensive mineralization of the compound to carbon dioxide was also observed, with 76 to 91% of the maximum theoretical CO_2 yield achieved in 28 days. Ready biodegradation in the manometric respirometry test was shown to correlate with biodegradability of the compound in surface water samples. Rapid biodegradation of bisphenol A was measured in wastewater effluent, and in river waters and sediment/water microcosms prepared with samples taken from receiving waters downstream of bisphenol A manufacturing facilities. Half-lives for removal of the compound were on the order of 2 to 4 days. Results of these studies, as well as others examining biodegradability in wastewater simulation tests, indicate bisphenol A would be expected to exhibit short lifetimes in the environment.

PHA094 Decolorization of Reactive Azo Dye Red 2 Under Various Redox Conditions. Beydilli, M.I.*; Pavlostathis, S.G.; Tincher, W.C., Georgia Institute of Technology, Atlanta, GA. Laboratory experiments were conducted to evaluate the decolorization of the reactive azo dye Red 2 under different redox conditions. All methanogenic and anoxic assays were performed at 35°C in the dark, and aerobic assays were performed at room temperature. A methanogenic culture achieved more than 90% dye decolorization at dye concentrations ranging from 50 to 2000 mg/L. The oxidation-reduction potential (ORP) values in the batch methanogenic assays varied from an initial value of -196 mV to a final value of -251 mV. The rate of dye decolorization was a function of biomass concentration. An aerobic, activated sludge-derived culture amended with an initial dye concentration of 400 mg/L did not achieve any significant decolorization over the 7-days of incubation. When the aerobic, mixed culture was incubated in an oxygen-free reactor amended with 300 mg/L dye and a mixture of dextrin/peptone, significant decolorization was not achieved during the initial 2-days of incubation when the ORP value varied from +111 to -40 mV. However, as the culture ORP value decreased further and stabilized around -250 mV, fast decolorization of the dye was observed at a rate comparable to that achieved under methanogenic conditions. Therefore, as long as relatively low redox conditions were maintained, decolorization was achieved irrespectively of the type of the microbial consortium and the electron accepting conditions. In all three assays, dye concentrations in media controls did not change over the incubation period demonstrating that the observed dye decolorization was biologically mediated. Similar results were obtained with several reactive azo dyes. These findings have implications on the development of a biological system to be used for the decolorization and reuse of reactive dyebaths in the textile industry.

PHA095 Comparison of Trichloroethylene biodegradation in the aerobic rhizosphere created by *Typha latifolia* vs. the anaerobic zone of wetland sediments - a microcosm study. Lingle*, D. M. and Bailey, F.C., Middle Tennessee State University, Murfreesboro, TN 37132. Freshwater wetlands can act as sinks for contaminants from surface runoff and groundwater. Wetland plants can play an important role in biodegradation of certain contaminants through absorption and the creation of an aerobic environment in their rhizospheres. Trichloroethylene (TCE) is a ubiquitous contaminant in many aquifers in the U.S. that is subject to microbial transformation under aerobic and anaerobic conditions. In this study, the potential for enhanced degradation of TCE in anoxic wetland sediments vs. the aerobic rhizosphere of *Typha latifolia* was examined. Glass microcosms (10"X14"X1") were constructed and experiments were implemented which involved plants vs. "no plants", sterilized vs. unsterilized sediment, and TCE (28.6ppm in sediment) vs. no TCE treatments. After 23 days samples were taken from the aerobic rhizosphere and analyzed for TCE concentration and compared with samples taken from the surrounding anaerobic sediment. "No plant" treatments involved only an anaerobic sediment sample. A significant ($F=45.242$; $\alpha<0.01$) difference was found between the TCE concentration in the aerobic rhizosphere sediments and anaerobic sediments of plant treatments ($X_{aerobic}$= 5.7ppm, $SE_{aerobic}$= 0.67; $X_{anaerobic}$= 24.7ppm, $SE_{anaerobic}$ = 3.98), indicating enhanced degradation of TCE in the rhizosphere of *Typha*. There was no statistical difference in TCE concentration between autoclaved and unautoclaved sediments, or in the anaerobic zone of plant treatments and "no plant" treatments. TCE metabolite formation in these systems is currently being studied.

PHA097 Reductive Dechlorination and the Intrinsic Bioremediation of Chlorinated Hydrocarbons in Wetland Systems. McInnis, D. L., Toxicological & Environmental Associates, Inc., Baton Rouge, LA; Kirschner, L. E., Ph.D., Toxicological & Environmental Associates, Inc., Baton Rouge, LA; Green, D., Directorate for the Department of Safety, Health and Environment (DSHE), United States Army – Aberdeen Proving Ground, MD; Wrobel, J., Directorate for the Department of Safety, Health and Environment (DSHE), United States Army – Aberdeen Proving Ground, MD; Pardue, J. Ph.D., Department of Civil and Environmental Engineering, Louisiana State University, Baton Rouge, LA; Williams, R., NPC Services, Inc., Baton Rouge, LA; Martino, L., Argonne National Laboratory, Washington, D.C.. Natural Attenuation studies have been performed at six Aberdeen Proving Ground (APG) sites located at Aberdeen Proving Ground, Edgewood, Maryland (Cluster 13, Grace's Quarters, J-Field, Nike Site, Southern Bush River, and Canal Creek). Similar studies have also been performed at the Petro Processors Superfund site located in Baton Rouge, Louisiana. Groundwater chemistry data show that a variety of microbially mediated redox processes including oxygen reduction, nitrate reduction, iron reduction, sulfate reduction, and methanogensis occur in the groundwater aquifers at each of these sites. The data indicate that strong methanogenic conditions predominate near the weland areas and are surrounded extensively by sulfate reducing conditions. Immediately adjacent to these sulfate-reducing areas, there is evidence of iron reducing conditions. Further up-gradient toward the former source areas, nitrate-reducing conditions begin to be predominated. There are at least three transitional redox zones along the flowpath downgradient from the former source areas. These are due to apparent transport of the geochemical parameters along the groundwater flowpath. Areas of oxygen reduction are occurring along the periphery of the plume(s). Chlorinated ethenes and ethanes from the former source areas are therefore first exposed to iron-reducing conditions and then to progressively more (sulfate reduction and methanogenesis) conditions downgradient near the wetland areas. These conditions strongly favor the natural degradation of chlorinated ethenes and ethanes at each site. Estimates of the biodegradation kinetics indicate that the half-lives of chlorinated compounds at the sites average approximately one to five years and less than one year near the marsh areas. This data indicates that natural attenuation is occurring at rates that have restricted plume migration to off-site receptors, thereby reducing potential health risks to human and ecological receptors. Acridine orange and epifluorescent direct cell counts (AODCs) were conducted at each of these sites and support the third line of evidence for natural attenuation of the chlorinated ethenes and ethanes should be considered as viable remedial alternatives at each site.

PHA098 Use of Epifluorescent Direct Cell Counts for Development of the Third Line of Evidence Supporting Natural Attenuation. Kirschner, L.E., Ph.D.*, Toxicological & Environmental Associates, Inc., Baton Rouge, LA; McInnis, D.L., Toxicological & Environmental Associates, Inc., Baton Rouge, LA. The three lines of evidence that are used to support natural attenuation of BTEX and chlorinated organic compounds in soil and groundwater are: (1) measurable decreases in the parent compound(s) and increases in concentrations of daughter compounds; (2) reductive changes in selected redox ion pairs; and (3) demonstration of increased microbial activity in the area of parent compound diminution. While microcosm studies provide useful information on the ability of indigenous microorganisms to degrade target compounds, time and funding for such studies is not always available. Therefore, alternate methods for demonstrating increased microbial activity within contaminant plumes becomes necessary. Direct cell counting techniques can provide useful data in a timely manner to support the third line of evidence for natural attenuation. To demonstrate this procedure, groundwater microflora from several locations were enumerated using epifluorescent direct cell counting techniques. The efficiency of two buffers for dissociating cells from soil particles and detritus were compared, and two fluorescent dyes (acridine orange and SYTO 16 (Molecular Probes, Eugene, OR)) were compared for ease of use, brightness, specificity, background staining characteristics, and cost. Data from the enumerations were used to demonstrate changes in microbial cell numbers, according to the sampling location with respect to the contaminant plume. Results indicate that microflora were present in high concentrations around the contaminant plume. These results indicate that epifluorescent direct cell counts can provide useful information to support natural attenuation. The methods are fast, cost effective, and provide support for the third line of evidence in support of natural attenuation.

PHA099 Combining Active and Passive Techniques for the Remediation of an Acrylonitrile Release in the Subsurface. Huntsman, B.E.*, Terran Corporation, Beavercreek, OH; Staples, C.A., Assessment Technologies, Inc., Fairfax, VA; Boyle, T.F. Solutia, Inc., Decatur, AL. A failure in a buried product pipeline resulted in the release of acrylonitrile (AN) to a silty, clay rich unsaturated soil and ultimately to the underlying groundwater. Following a detailed hydrogeologic investigation to define the concentration, rate and extent of AN movement in the subsurface, a two-phase remediation program was implemented. The first phase consists of active removal of the highest concentrations of AN using extraction wells and off-site treatment of discharge water. The second phase passively monitors the effectiveness of natural attenuation of residual AN concentrations and provides necessary information to adjust the active removal system to insure minimal product migration. Periodic monitor well network sampling with analyses for AN, acrylamide, acrylic acid, acetic acid, propionic acid as well as other indicator parameters has confirmed consistent degradation of AN. Threshold concentrations below which significant AN degradation is initiated and the decay half life variability of AN and degradation daughter products in groundwater were determined. In less than two years, a five-fold decrease in AN groundwater concentrations have been documented. AN degradation rates are compared and contrasted to previously published rates obtained from laboratory studies. Difficulties in extrapolating laboratory data to observed natural attenuation of AN under field conditions is evaluated through the development and use of analytical models.

PHA100 Control of pH with Encapsulated Buffer During Denitrification in Subsurface Soils. Rust, C.M.,* Aelion, C.M., and Flora, J.R.V. University of South Carolina, Columbia, SC. In anaerobic groundwater and saturated soils, denitrification is often a dominant respiratory process. This process can cause pH fluctuations in the solution surrounding a bacterium. If the pH becomes too high as a result of denitrification, or too low due to the production of CO_2 or HCl from contaminant degradation, microbial metabolic activities may slow or cease. The objective of this research is to investigate the control of pH in soil columns using microencapsulated phosphate buffer. Soils obtained from the Savannah River Site were tested in 125 ml microcosms for their ability to denitrify under anaerobic conditions. The acetylene block technique was used measure the production of nitrous oxide from the reduction of nitrate. Headspace sampling of N_2O over time was measured on a gas chromatograph equipped with an electron capture detector. The pH of the microcosms before and after the addition of 1 mg N/g dry soil KNO_3 was measured with an Orion ROSS™ electrode in water and 2M KCl extracts. Biotic controls showed minimal denitrification compared to treatment vials which yielded up to 4213 nmol N_2O-N/g dry soil in the headspace after 5 days of incubation. pH increased from 5.4 to 7.9 in treatment vials. The microcapsules (1mm diameter) are inserted into soil columns and when the pH in the column rises above 7.0, the pH-sensitive polymer coating melts and the buffer dissolves into the nearby solution, allowing the pH to return to neutral. Preliminary experiments carried out with the encapsulated buffer in a batch bioreactor showed they were effective in maintaining a neutral pH within 0.2 pH units. The pH in controls rose to 9.0. These results indicate that encapsulated buffer may be effective in maintaining microbial degradation activity and pH control in flow-through soil columns.

PHA101 Bioavailability and Elimination of the Fluoroquinolone Antibiotic Ciprofloxacin in Wastewaters in a modified OECD 302B assay. Hartmann, A.[1]*, Alder, A.C.[2], Koller, T.[1], Widmer, R.M.[1]; [1] Swiss Federal Institute of Technology (ETH), CH-8092 Zurich, Switzerland and [2] Swiss Federal Institute for Environmental Science and Technology (EAWAG), CH-8600-Dübendorf, Switzerland. Low-level effects of environmental pharmaceuticals are under vigorous debate. Drugs often enter the aquatic environment via wastewaters. Our aim was to investigate the bioavailability and elimination of Ciprofloxacin (CIP, the leading human fluoroquinolone antibiotic) during wastewater treatment. CIP was shown to occur in hospital wastewaters at 5 to 100 µg/L by reversed-phase (RP) HPLC. As a model system, a modified OECD 302B assay was used. Native or spiked hospital wastewaters were treated for up to 24 days using activated sludge concentrations of 0.1 and 1 g dry matter/L (d.m./L)., respectively. Initial CIP concentrations were 5 to 184 µg/L. Sample analysis after 0.45 mm filtration included DOC for monitoring the activated sludge elimination capacity, dissolved CIP (by RP-HPLC) and CIP bioavailability (using a highly sensitive bacterial genotoxicity assay with a LOEC of 5 µg/L for CIP), the umuC test. After 24 days we found CIP to be eliminated to 56 ± 11% with 0.1 g d.m./L activated sludge and to 85 ± 1% with 1 g d.m./L activated sludge concentration. After 3 hours, just 4 ± 14% (0.1 g d.m./L sludge) and 18 ± 9% (1 g d.m./L sludge) of the initial CIP was eliminated. This strongly indicates that elimination was not exclusively due to sorptive effects. DOC removal was unaffected even at the highest CIP concentrations. Genotoxicity essentially paralleled the RP-HPLC results indicating a substantial bioavailability of CIP. Our results could be evidence for a different behaviour of fluoroquinolone antibiotics in wastewaters compared to soils or sediments, where strong sorption and low bioavailability are reported.

PHA102 Kinetic data from degradation screening tests: Do they reflect the situation in river waters? Geisel, U., Steber, J., Henkel KGaA, Düsseldorf – Germany. The Environmental Risk Assessment process of new and existing chemicals is based on the prognosis of the environmental concentration (PEC, predicted environmental concentration) and the ecotoxicological no effect concentration (PNEC). In the context of the exposure analysis kinetic data of the elimination in river waters may play a crucial role for the evaluation of the PEC. Elimination rate constants and half-lifes are measured by river monitoring or examinations in river models. As these examinations are very time and money consuming, deriving these data from screening tests would be a useful alternative. We present kinetic elimination data of selected readily biodegradable substances obtained from river water screening tests and compare these with data from river models and river monitoring examination.

PHA103 Development of Draft National Default Bioaccumulation Factors (BAFs) for 28 Organic Chemicals. D. O. McIntyre, W. H. Clement and T.K. Linton, Great Lakes Environmental Center, Columbus, OH, K. Sappington, U.S. EPA, Washington D.C., and L. Burkhard, U.S. EPA, Duluth. The U.S. EPA intends to publish a *Federal Register* notice proposing revisions to the Agency's ambient water quality criteria methodology for the development of human health criteria. The proposed revisions contain a national BAF methodology component, based largely on the Great Lakes Water Quality Guidance BAF methodology. The U.S. EPA is advocating that States and Tribes update their existing ambient water quality criteria using EPA's revised methodology. This poster presents national default baseline BAFs for 28 organic chemicals (24 halogenated hydrocarbons and 4 PAHs) and outlines the process of how they were developed. A baseline BAF is the ratio of the concentration of a substance in lipid-normalized tissue to its freely dissolved concentration in the surrounding water in situations where the organism and its food are exposed. As a result, this expression accounts for the effects of lipid content in the aquatic organism and the DOC and POC in the water column on bioaccumulation. A literature search identified approximately 2000 articles that potentially contained relevant information for BAF development. A review of the abstracts reduced the list of potentially relevant articles to 600 which were retrieved and reviewed for extraction of data usable for BAF development. Data extracted from the literature were used to develop baseline BAFs using four methods: 1) directly from field-measured BAFs, 2) predicted from biota-sediment accumulation factors (BSAFs), 3) predicted from a laboratory-measured bioconcentration factor (BCF) and a food chain multiplier (FCM), and 4) predicted from a K_{ow} and a FCM. The U.S. EPA intends to use these national default BAFs to revise EPA's national ambient water quality criteria recommendations for 28 chemicals.

PHA104 Cooperative Natural Resource Damage Assessments: Can We Walk the Talk? Ammann, M.J. Chevron Research and Technology Company, Richmond, CA.Now that the legal briefs challenging the National Oceanographic and Atmospheric Administration's natural resource damage assessment (NRDA) rule have gone through the spin dry cycle of our judicial system, attention is being focused, by trustees and industry representatives alike, on how to live with the final rule. Instead of "See you in court", the mantra chanted now is "cooperative assessment". Working cooperatively is indeed the preferred path. But what does it mean to conduct a cooperative assessment? How do we do it? How do we measure our success at working cooperatively through a process that, even under the best of circumstances, can test the patience of Job?The author takes a stab at outlining some of the philosophical considerations and challenges that the people in the NRDA trenches have to confront to make cooperative assessments a reality. For example, what is the role of the scientists working on the assessment? What is the interplay of science, experience, and good judgment? What are the characteristics of a cooperative assessment?

PHA105 Comparative Assessment of Point and Non-point Source Impacts on Invertebrate Communities. M. Eversen*, T.W. La Point, J. Morse, C. Lee, Clemson University, Clemson, SC, USA and Texas Tech University, Lubbock Texas, USA. Non-point source runoff during storm events presents a continuous episodic dose of heavy metals and petroleum byproducts. The Reedy River in Greenville South Carolina is a prime example of a river with exposure to both point and non-point source contamination. The Reedy was the site of a pipeline rupture carrying diesel fuel, releasing nearly one million gallons of fuel into the river in June of 1996. This insult compounded the impact of continuous non-point contamination from urban runoff. The amount of area that is paved continues to increase with rapid development, as does the storm surge following rain events. The goal of this study was to assess the relative impact of the diesel pipeline rupture, urban runoff contaminant load and storm surge on Reedy River invertebrate communities. Preliminary results from *C. dubia* tests failed to demonstrate toxicity despite the measure of 105 to 238 ppb semi-volatile polyaromatic hydrocarbons in the tested storm effluent. The invertebrate surveys done over two sampling periods with artificial substrates indicate a distinct repeated correlation between habitat and invertebrate community health. In both survey the number of taxa , quality of the habitat, and percentage macroinvertebrate population that are filter feeders significantly declined at sites located in and below downtown Greenville. Conversely, the tolerance index or FBI, Family Biotic Index, increased with the distance downstream. In all cases sedimentation, resulting from bank erosion during storm surges, and loss of riparian buffer zones to development, appear to be the primary cause of poor invertebrate community structures at downstream sites rather than toxicity of point and non-point source contaminants.

PHA106 Ecological Risk Assessment as a Decision Tool for Remediation of Contaminated Sediments at Great Lakes Areas of Concern. Passino-Reader, D.R.*, Hickey, J.P., USGS, Ann Arbor, MI. Contaminated sediments are the primary cause of impairment of beneficial uses of the 43 Areas of Concern (AOCs) in the Great Lakes in the US and Canada. Remediation of contaminated sediments requires information on the degree of exposure and effects on fish, wildlife, and other aquatic biota. A mixture of contaminants and other stressors at these river, harbor, and nearshore areas are present and impair ecosystem health. We conducted ecological risk assessments for selected fish and benthic macroinvertebrates at the Buffalo River, NY, Saginaw Bay, MI, and Grand Calumet River/Indiana Harbor, IN, which are sites for which assessments were conducted under the USEPA ARCS project. We examined trends of contaminants in sediments and the structure of the fish and benthic communities with time to provide evidence of natural remediation processes and of restoration of biotic communities from remedial actions. Probabilistic models of ecological risk assessment were developed to provide decision makers with tools for selecting remediation alternatives. Uncertainties were examined by Monte Carlo analysis; however, greater consistency in design of monitoring efforts is needed for application of some distributions. The three AOCs give evidence of decreased contaminant levels and improved abundance and structure of fish and benthic communities resulting from restoration efforts and reduced input from contaminant sources.

PHA107 Comparison of Natural Recovery of Surface Waters from Coal and Phosphate Mining. Grippo, R.S.*, Arkansas State University, State University, AR; Pratt, J.R., Portland State University, Portland, OR. The extraction of minerals by mining often has serious environmental consequences on aquatic ecosystems. Regulations directed at the development and implementation of plans for recovery of mine-impacted aquatic systems lag behind those required for impacted terrestrial systems. This presentation provides an overview of current information on the natural, long-term recovery of aquatic systems from impacts associated with coal and phosphate mining. We compare the physico-chemical and ecological characteristics (primary productivity, species diversity/richness) of surface waters impacted by both mining types and how these factors affect recovery. A specific case history of physico-chemical and ecological recovery from each type of mining is presented. The methods used for evaluating recovery in each case history provide some examples of the different approaches needed to characterize recovery in both lentic and lotic systems. We conclude with specific recommendations for evaluating mine recovery, including establishment of sampling strategies useful for determining intermediate levels of recovery, measurement of ecosystem structure to evaluate recovery, use of toxicity based water quality standards rather than performance standards for evaluating mine effluent and working towards a goal of directed ecosystem replacement.

PHA108 Recovery Potential of Parker Run from Intermittently Toxic Coal Mine Effluent and a Coal Slurry Spill, Meigs County, OH. Latimer, H.A.*, Cherry, D.S., Currie, R.J., Babendreier, J.E., Virginia Polytechnic Institute and State University, Blacksburg, VA and J.H. Van Hassel, American Electric Power Company, Columbus, OH. Effluent from Meigs #31 coal mine in Meigs Co., Southeastern, OH, discharges into Parker Run, a tributary of Leading Creek, Ohio River drainage. During periods of low upstream dilution the effluent comprises ~ 100% of the flow in Parker Run and >90% of flow in the mainstem of Leading Creek. The mine effluent is characterized by high conductivity (5,500 - >6,000 μmhos/cm), high total dissolved solids (TDS, 3,300 – 6,000 mg/L) elevated pH (8.3+), and high ion concentrations (sulfate ~ 2,000 ppm, sodium ~ 1,000 ppm and chloride ~ 330 ppm). The water periodically is acutely toxic as 48 hr *Ceriodaphnia dubia* LC_{50} values ranged from 66 - >100%. Acute toxicity is accompanied by conductivity in excess of 6,000 μmhos/cm, and was hypothesized to be a toxic threshold for either sodium or TDS concentrations in the effluent. *Corbicula fluminea* in-situ (35-day) toxicity testing has shown Parker Run to be chronically toxic, and under low dilution, toxicity extends into the Leading Creek mainstem. Although benthic macroinvertebrates in Parker Run are recovering from an April 1997 coal slurry spill, recovery was impeded by the intermittently toxic nature of the effluent. Immediately after the spill, macroinvertebrate assemblages in Parker Run ranged from 0-10 in abundance at six sampling stations. Five and 9 months later, some recovery was observed by increasing richness while total abundance remained depressed. The intermittent toxicity of the effluent appeared to correlate strongly to acutely toxic levels of sodium and TDS.

PHA109 Ecotoxicological Monitoring at a Remediated Superfund Site B. Anderson*, J. Hunt, B. Phillips, R. Tjeerdema, University of California, Santa Cruz; R. Fairey, San Jose State University; M. Martin, California Department of Fish and Game. The United Heckathorn Superfund Site in Richmond California was polluted with DDT and dieldrin as a result of pesticide processing activities from 1947 through 1966. As part of the remediation activities at this site, contaminated sediment was removed via dredging from Lauritzen Channel, an adjacent marine habitat. The dredged channel was then capped with clean San Francisco Bay sand in an effort to restore benthic habitat. A monitoring program was designed to assess whether remediation effectively reduced concentrations of chemicals of concern in the marine environment to levels that prevent risk of injury to biota. Monitoring included three components: measures of ambient concentrations of chemicals of concern in both the sediments and overlying water; measures of bioaccumulation using field-deployed bivalves; and laboratory toxicity assessments using field collected sediment and water samples. Monitoring was conducted immediately prior to and after dredging and capping of the site. Results of studies prior to remediation indicated chemical contamination by DDT and dieldrin, solid-phase toxicity to amphipods, toxicity to mollusc larvae exposed at the sediment-water interface, and bioaccumulation of DDT and dDieldrin in caged bivalves. Post-remediation monitoring indicated that bioaccumulation of DDT and dieldrin was reduced in caged bivalves, although some chemical residue remained. In addition, although bivalve larval development at the sediment water interface improved, amphipod survival declined significantly in post-remediation sediments. Post-remediation benthic community structure indicated a transitional benthic community. These results will be discussed relative to post-remediation changes in water column and sediment chemical contamination, and with respect to the overall goals of the remediation program.

PHA110 Sediment Metal Concentrations Before and After a Marine Dredging Operation. Rudis, D.D., U.S. Fish and Wildlife Service, Juneau, AK; McNutt, C.D., Jr, U.S. Coast Guard, Juneau, AK. A preliminary investigation of a marine ways facility in Ketchikan, AK, determined that past disposal practices had resulted in marine sediment contamination by metals. Releases of contamination to the intertidal zone were primarily from boat repair activities and battery storage. Sediment and biota samples were collected both before and after removal activities to determine if metal concentrations were reduced after sediment removal. Sediment, mussel, and fish samples were collected from the Ketchikan facility and a reference location. Chromium, copper, lead, nickel, and zinc concentrations were elevated in mussels. Dredging activities removed sediment using clam and airlift dredges. The minimum amount of sediment removed was estimated at 400 to 600 cubic yards. Resampling was conducted eight months after removal activities. Sediment samples collected did not show a significant change in metal concentrations, and most metals (As, Cr, Cu, Hg, Ni, Pb, Zn) were detected in higher concentrations. However, mussel tissue samples had lower mean concentrations of Cd, Cu, Pb, and Zn. Comparison with Washington State Sediment Management Standards indicate As, Cu, Hg, and Zn exceed guidelines and As, Cu, Pb, and Zn are at NOAA ERM values. Based on these results, removal operations appeared to have resuspended fine-grained sediments resulting in an additional contaminant release.

PHA111 Where is the Environmental Relevance in the SETAC Life Cycle Impact Assessment Normalization Step? Dubreuil, A., Natural Resources Canada, Ottawa, Ontario SETAC North America and SETAC Europe are developing the methodology of life cycle impact assessment. That work is having a considerable impact internationally as the structure of the ISO/CD 14042.3 –Life Cycle Assessment—is similar to the SETAC framework. According to the SETAC framework, the LCIA directs the inventory data collection according to the goal of the study. The classification step assigns the life cycle inventory results into impact categories. The characterization step uses specific environmental models and each inventory in a given impact category is converted into category equivalents which are aggregated into a category indicator. The normalization step divides the category indicator by a reference value. The valuation step involves a formal ranking (weighting factor) across impact categories of the normalised results to generate a single number (score). One of the aims of normalization is to help communicate results. The normalised results are used in the valuation step. Commonly normalization is carried out by dividing the category indicator of the functional unit by the total category indicator for the region under consideration. As a consequence of this model, for a constant weighting factor, a reduction in the total emission for a given geographic area in function of the time leads to an increase in the score per functional unit of emission. As the emissions in the given geographic area can change significantly in function of time, the valuation step needs to be changed accordingly. Therefore there is a need to have more robust normalization approaches which are more environmentally relevant and which reduce the necessity to review the weighting factor in the valuation step. An improved normalization step will be discussed.

PHA112 Total Cost Accounting (TCA): Integrating the Business and Product Life Cycles. V. L. Morris, Arthur D. Little, Inc., Cambridge, Massachusetts. Under the sponsorship of the American Institute of Chemical Engineers, a task force of 15 multi-national companies (chemical, pharmaceutical and pulp & paper) was formed (with Arthur D. Little as the study contractor) to develop a standardized methodology for Total Cost Accounting (TCA). This task force is motivated to better understand their environmental costs and risks in order to sustain competitive advantage. Globally, governments are imposing harsher regulations, while other parties, including customers, shareholders, suppliers, consumer groups, environmental organizations and even banks and insurance companies are formulating new requirements with respect to the environmental impacts of products. During the initial phase of the program, an international survey of the best industrial practices currently employed in TCA was conducted. Also surveyed were the automated tools and databases that are available for this purpose. The results of the survey showed that, internationally, there is a real expressed need to better understand and manage the environmental costs and risks associated with industrial processes, but organizations do not have the internal systems or tools to accomplish this goal. Using an iterative process, whereby the best practices and tools were reviewed and "tested" by these 15 companies, an approach to a methodology was developed. The methodology will employ a streamlined life-cycle approach to identify and quantify environmental impacts and costs. The integration of the life cycle and business cycle impacts of the products will yield a decision support tool to make better informed decisions regarding new product and process development, as well as modifications to existing products and processes. Leading edge automation tools are being developed, primarily in Germany, to facilitate financial inventories, impact assessments and improvement assessments. The real benefit of this program is the willingness of industrial competitors to collaborate and develop standards and a methodology that is widely applicable to a broad range of companies. Unlike other programs, where the technology developer designs a product that is believed to meet customer needs, this program has utilized the expressed needs of industrial organizations to develop a product designed for their specific operations and is being validated by them in real-time.

PHA113 Life Cycle Assessment (LCA) - A Guide to Approaches, Experiences and Information Sources. A.A. Jensen*, L. Hoffmann, B.T. Moeller, and A. Schmidt, dk-TEKNIK Energy and Environment, Søborg, Denmark, J. Elkington, and F.van Dijk, SustainAbility Ltd, London, U.K., K. Christiansen, Sophus Berendsen A/S, Søborg, Denmark and I. Andersson, European Environment Agency, Copenhagen, Denmark. In order to increase the accessibility of environmental management tools, the European Environment Agency (EEA) has produced several environmental management guides of which the most recent covers life cycle assessment (LCA). The guides are produced both as paperbacks and as internet files located on the EEA homepage (http://www.eea.eu.int). The LCA guide contains a brief history of LCA and its potential role in sustainable development. Different applications of LCA such as product development, marketing and ecolabelling are discussed, and the methodological framework of LCA is explained based on the recent ISO and SETAC developments. The Guide also contains a lot information about LCA newsletters, journals, books, reports, software, the most important internet addresses, and a database of about 200 organisations, mainly European, involved in LCA practise and development. The internet version has links to other LCA websites and e-mail addresses.

PHA114 Life Cycle Assessment of a Nortel Telephone Supported by Environment Canada. Brickman, L., Ecobalance, Inc., Rockville, MD; Coulon, R.*, Ecobalance, Inc., Rockville, MD; Noble, D., Nortel, Ottawa, ON, Canada. The aim of this joint study between Environment Canada and Nortel is to carryout a Life Cycle Assessment (LCA) on a manufactured product in order to identify improvement strategies. The main benefit expected from this LCA study is the demonstration of the concept of product Ecodesign on LCA. Through the analysis of several improvement scenarios, this LCA will provide a case study of how life cycle assessment can be applied to design more environmentally thoughtful products. This study will provide a clear environmental picture of the current design of the telephone. The functional unit is the Northern Telecom M7310 Business Telephone over its life span which includes refurbishings. The telephone set is not self-sufficient and a Key System Unit (KSU) central system to which more than one phone is attached must accompany it. The study of this KSU system is taken into account as a scenario analysis (to put the phone results in perspective) and not as a part of the functional unit itself. Other prospective scenarios analyzed include a new integrated keypad technology, thin-wall housing, and alternate end-of-life strategies. This paper marks the first time that results of this study will be published in a public manner. It was found that in terms of the greenhouse gas effect and air acidification, the use phase dominates the life cycle. Within the manufacturing phase, the burden from the main electronics subassembly dominates these life cycle indicators. The copper in the phone cords contributes the most to the raw material depletion index, and the production of paper and labels provides the most potential burden to eutrophication effects.

PHA115 Interfaces in Environmental Chemistry and Life Cycle Impact Assessment – A Case Study: Solvent Emissions in Textile Dry Cleaning. Mössner, S.G.*; Flückiger, P.H.; Weidenhaupt, A.[1]; Hungerbühler, K., Laboratory of Chemical Engineering, Safety & Environmental Technology Group, Swiss Federal Institute of Technology, Zürich, Switzerland; [1] Centre de Ressources des Technologies pour l'Environnement, Esch-sur-Alzette, Luxemburg. Significant amounts of various chemical solvents used in textile dry cleaning - perchloroethylene (PER) and hydrocarbons (HC) - are emitted into our environment, especially the atmosphere. In order to achieve an overall environmental assessment of solvent emissions within Life Cycle Assessment (LCA) and Product Risk Assessment the environmental chemistry, the metabolites (e.g., trichloroacetic acid, tetrachloromethane, carbonyl compounds, peroxyacyl nitrates), and the resulting environmental impacts must be included. So far this has not been done systematically. In this study, degradation pathways of PER and HC emissions in air, water, and soil are identified and evaluated. Existing classification factors for PER and HC are compared with new calculated classification factors generally used in LCA. The new approach adds significant information so that decision-making results can be affected. Approaches for the scientifically adequate handling of uncertainties occurring within environmental chemistry and environmental impact assessment are proposed. Finally, an environmental assessment and comparison of PER and HC is provided.

PHA116 An Investigation of Dynamic Economic Input-Output Modeling to conduct Dynamic Life Cycle Assessment studies. Gloria, T. P.*, and Levine, S. H., Tufts University, Medford, MA. This investigation examined the application of an established dynamic economic interdependency modeling methodology to address the inherent shortcomings of static analysis in the existing conventional LCA methodology. The methodology applied was the dynamic economic input-output model defined by Leontief, W., Duchin, F. and Szyld, D. Specifically, this study explored the LCA of an electric vehicle (EV), that seeks to answer the following question: What is the net increase or decrease in materials use, energy use, Toxic Release Inventory (TRI) emissions, "criteria" air emissions, and green house gases by introducing Lithium-Polymer battery powered Electric Vehicles (EVs) in the U.S. economy for a period of 10 years, 1998 to 2008? The research resulted in the successful demonstration of a methodology to conduct dynamic LCA studies utilizing the dynamic input-output framework. Emphasis was given to developing a methodological approach to incorporate technological change as it effects environmental impacts in a time varying non-linear dynamic analysis.

PHA117 Temporal Trends in Environmental Concentrations of Tributyltin Observed in the United States 1982–1997. Keithly, J.*, Brancato, M.S., Parametrix, Inc., Kirkland, Washington. Concern over release of tributyltin (TBT) in aquatic environments and its impacts on non-target organisms arose in the early 1980s, which led to the passage of the Organotin Antifouling Paint Control Act (OAPCA) in late 1988, limiting its use. As a result, national programs were initiated to evaluate the effectiveness of the legislation by monitoring the status and trend of TBT in U.S. coastal environments. The programs in the U.S. include those conducted by (1) the U.S. Navy, (2) a consortium of TBT manufacturers as required by the U.S. EPA, and (3) NOAA's National Status and Trends Mussel Watch project. Cumulatively these programs have monitored the trends of TBT concentrations in U.S. coastal environments for over 14 years. The regions studied include the Pacific, Atlantic, and Gulf coasts, and the Great Lakes. Matrices monitored include water, sediments, and bivalve tissues in areas of concern, such as industrialized areas, as well as in fish/shellfish habitats and open coastal waters. The data from the national monitoring programs show an average decrease of TBT concentrations of 67% in surface waters, 62% in sediments, and 39% in bivalve tissues. Overall, both the national monitoring programs and regional studies show generally low concentrations of TBT in the water column, with a decreasing trend in all matrices in U.S. coastal environments since the passage of OAPCA.

PHA118 Assessment of Risks to Otters Feeding on Bivalves Containing Tributyltin. Cardwell. R.D.*, Brancato, M.S., Keithly, J., Parametrix, Inc., Kirkland, Washington. Recent studies have postulated that tributyltin (TBT) and other butyltins may have contributed to the deaths of sea otters off the California coast. TBT is a suspected contributor to the deaths of these animals as it bioaccumulates in bivalves, a common sea otter food item. Potential risks to sea otters were assessed using U.S.TBT bivalve data probabilistically by fitting distributions of Probability Distribution Functions (PDFs) to the exposure concentrations and other exposure parameters, such as food ingestion rate and body weight. Although no TBT toxicity data for sea otters were available, TBT toxicity data (e.g., no observed adverse effects levels and lowest observed adverse effects levels) for surrogate wildlife species were used after applying an appropriate uncertainty factor following EPA guidance. The potential for risk to sea otters was considered if the estimated dose exceeded the toxic dose. This was calculated by integrating the exposure and effects PDFs to estimate sea otter individuals in a population that may be affected. The results of these comparisons will be presented.

PHA119 Cost Analysis of TBT Self-Polishing Copolymer Paints and Tin-Free Alternatives for Use on Deep-Sea Vessels. Damodaran, N., Pendleton, M. Mulligan, C. and Felmy, J., Princeton Economic Research, Inc., Rockville, MD and Toll, J., DeForest, D. and Kluck, M.J., Parametrix, Inc., Kirkland, WA. We conducted a comparative analysis of the costs of tributyltin (TBT) self-polishing copolymer (SPC) antifouling paints and their alternatives, to better understand the economic impact of restricting the use of TBT on deep-sea vessels. Costs evaluated include antifouling paint costs, dry-docking rates, clean hull fuel consumption, and fuel consumption penalties as a result of hull fouling. TBT SPC paints offer significant cost savings to ship owners and operators, because their five-year dry-docking interval reduces dry-docking costs and revenues lost during dry-docking, assuming a 30-month dry-docking interval for tin-free antifouling paints. As a conservative estimate, we assumed no fuel penalty in comparing TBT SPCs and tin-free SPCs. Tin-free SPCs are 95 to 146% more expensive, and copper ablatives are 156 to 401% more expensive than TBT SPCs. This is due to higher dry-docking costs, revenues lost, paint costs, and, in the case of copper ablatives, fuel costs. We have conservatively assumed that TBT and tin-free SPCs provide the same fuel efficiency, so the environmental risks or benefits of switching from a TBT SPC to a tin-free SPC would have to do primarily with the pesticidal impacts on non-target organisms. What data are available suggest that tin-free antifouling paints do pose risks to the environment. None of the tin-free alternative antifouling paints have been shown to be as safe as or safer for the environment over the long term than TBT SPC paints for use on deep-sea vessels.

PHA120 Tributyltin in Market-Bought Seafood from Europe, Asia, and North America and Estimated Human Health Risks. Cragin, D.W.*, Farr, C.H., Elf Atochem North America, Inc., Philadelphia, Pennsylvania; Cardwell, R.D., Keithly, J.C., Henderson, D., Robinson, S., Parametrix, Inc., Kirkland, Washington. This study assessed potential human health risks posed by tributyltin (TBT) in seafood purchased from seafood markets in Europe, Asia, and North America. In August and September 1997, samples of fish, crustaceans, cephalopods (i.e., squid), and bivalve mollusks were purchased at local markets in three European cities, three Asian cities, and two North American cities. Concentrations of TBT in samples were determined and used to calculate potential human health risks from consumption of market-bought seafood. Tributyltin concentrations compared well with previous data (1989) collected in the United States. The assessment indicated health effects from exposure to TBT continue to be unlikely from consumption of market-bought seafood, even under worst-case conditions. No risk to human health was predicted even when accounting for the uncertainties in consumption rates and in the toxicity of TBT.

PHA121 Butyltins in Fishes and Sediments of the Southern Baltic Sea. Duda, C.A.*[1], Senthilkumar, K.[2], Villeneuve, D.L.[1], Kannan, K.[1], Falandysz, J.[3], and Giesy, J.P.[1]. [1]Michigan State University, East Lansing, MI; [2]Ehime University, Matsuyama, Japan; [3]University of Gdansk, Gdansk, Poland. Concentrations of mono- (MBT), di- (DBT), and tri- (TBT) butyltin were measured in eggs, liver, and muscle of twelve fishes from four regions of the Baltic Sea - the Firth of Vistula, the Gulf of Gdansk, Puck Bay, and the Vistula River. Butyltins were quantified by gas-liquid chromatography with flame photometric detection (GC-FPD). The overall concentration ranges among all the fish sampled from the four sampling sites were: 3 to 79 ng/g for MBT; 6 to 1100 ng/g for DBT; 7 to 3600 ng/g for TBT; and 16 to 4800 ng/g for total BTs. Concentrations of total BTs in the Firth of Vistula, the Gulf of Gdansk, Puck Bay, and the Vistula River ranged from 78 to 4800 ng/g, 83 to 100 ng/g, 78 to 3300 ng/g, and 32 to 120 ng/g, respectively. The greatest concentration of total BTs was found in herring liver from the Firth of Vistula (4800 ng/g) and in roach muscle from Puck Bay (3300 ng/g), while the least concentration was found in burbot eggs and liver from the Vistula River (39 and 32 ng/g, respectively). TBT was the major compound among the BTs, suggesting the exposure of fish to recent sources of TBT. Sediment samples from the same region were analyzed for butyltin compounds and noticeable concentrations in the range of a few hundred parts per billion (ppb) were detected in areas near large harbors.

PHA122 Accumulation of Butyltin Residues in Southern Sea Otters Found Dead Along Coastal California. Kannan, K.*, Guruge, K.S., Thomas, N.J., Tanabe, S. and Giesy, J.P., Michigan State University, East Lansing, MI; U.S. Geological Survey, Madison, WI; Department of Environment Conservation, Ehime University, Japan. Tributyltin (TBT) has been used extensively since the 1960s as an antifouling agent in marine paint formulations to prevent the attachment of barnacles and slime on boat hulls and aquaculture nets. While a great deal of attention has been paid to investigate the accumulation and toxic effects of TBT and its breakdown products, DBT and MBT, in lower trophic aquatic organisms such as bivalve mollusks and gastropods, studies in higher trophic vertebrate predators were not available until 1995. Accumulation of butyltin compounds (MBT+DBT+TBT) has been reported in marine mammals such as dolphins. In this study, butyltin compounds were measured in liver, kidney and brain tissues of adult southern sea otters *(Enhydra lutris nereis)* found dead along the coast of California during 1992-1996. Hepatic concentrations of butyltin compounds ranged from 40 to 9200 ng/g, wet wt, which varied depending on the sampling location and gender. Concentration ranges of butyltins in sea otters were comparable to those reported in stranded bottlenose dolphins from the U.S. Atlantic coast during 1989-1994. Notable accumulation of butyltins in sea otters could be explained by their bottom-feeding habit, and the diet that consists exclusively of invertebrates such as mollusks and gastropods. Livers of female sea otters contained approximately 2-fold greater concentrations of butyltins than those of males. The composition of butyltins in sea otter tissues was predominated by TBT in most cases. Sea otters, which were affected by infectious diseases contained greater concentrations of butyltins in their tissues than those died from trauma.

PHA123 **The Present Status of Tributyltin Copolymer Antifouling Paints Versus Tributyltin-Free Technology.** Stewen, U.*, Gerigk, U., Schneider, U.Witco GmbH, Bergkamen, Germany. Approximately 70% of the world fleet are protected against fouling organisms by modern tributyltin (TBT)-based self-polishing copolymer (SPC) systems. There are ongoing discussions at the International Maritime Organization (IMO) about the harmful effects of the use of antifouling paints for ships. The search to develop alternative systems which completely satisfy technical and environmental demands has proved to be a complex problem. The mechanism of TBT-SPC systems will be clarified using surface sensitive techniques like XPS, AFM, and GA-FTIR. While the bioactive ingredient is chemically fixed to the polymer chain, a controlled and slow hydrolysis at the active zone of the paint surface occurs and guarantees a constant but very low TBT release from ship hulls. The detailed understanding of this unique mechanism to prevent fouling is the basis for the development of alternative systems. An overview is given about recently advertised new antifouling technologies, their mechanism, and potential environmental fate.

PHA124 **Risk assessment of copper as a marine antifoulant.** Karman, C.C*.; Jak, R.G., TNO Department for Ecological Risk Studies, Den Helder, The Netherlands; Kramer, K.J.M., Mermayde, Bergen, The Netherlands. Copper, as introduced as a marine antifoulant, could be hazardous for the environment when present in elevated concentrations. Copper is, however, an essential element for life, which implies that biota have developed active regulation mechanisms to control internal body concentrations within a certain range. Not all copper is present in a bioavailable form. There is sufficient evidence that the ionic form of copper, Cu^{2+}, is the dominant bioavailable chemical form in the dissolved phase. Lowest toxicity levels reported for Cu^{2+} are approximately 10^{11} M. Risk assessment studies for the marine environment on the basis of total copper concentrations may be considered too conservative. A risk assessment for marine areas on the basis of the ionic species, would be more relevant. Since there is not yet an analytical method available to measure Cu^{2+} concentrations, equilibrium concentrations of all copper species are usually estimated using a speciation model. The speciation is a function of several water quality parameters, and organic ligand concentrations and their binding capacities. In this study, the free copper concentration was estimated by using a speciation model for different marine environments (harbour, estuary, open sea), each with a relevant set of water quality parameters. It was identified which environmental factors contribute most to the binding of copper and at which total copper concentrations and water quality parameter values, enhanced copper levels might result in marine environmental risks. For risk assessment studies, it is recommended to apply speciation modelling to obtain free copper concentrations for both the exposure concentration and the toxicity threshold values.

PHA125 **Sources of Polycyclic Aromatic Hydrocarbons to Air, Water, and Sediments of Chesapeake Bay.** Dickhut, R. M.*, Canuel, E.A., Gustafson, K.E., Liu, K., Murray, K.Y., Walker, S.E., Edgecombe, G., Gaylor, M.O., MacDonald, E.H., Virginia Institute of Marine Science, College of William and Mary, Gloucester Point, VA Polycyclic aromatic hydrocarbons (PAHs) are carcinogens released into the environment largely by the incomplete combustion of fossil fuels. We collected over 200 air, 75 water, and 28 sediment samples, in addition to 66 rain and 32 sea surface microlayer samples from throughout the Chesapeake Bay region from 1991-1998, and analyzed these samples for various PAHs. Particulate PAHs in air, rain, and surface waters of Chesapeake Bay are estimated to be derived from both automotive and coal combustion sources; however, PAHs in surface sediments of the bay are estimated to be predominantly coal-derived. Consequently, PAHs input to the bay from automobiles are either degraded in the air and water prior to deposition to the sediments or are diluted by historically or recently deposited coal-derived PAHs in the seabed.

PHA126 **Ionizing Radiation-Induced Degradation of PCBs in Oil.** Chaychian, M, Al-Sheikhly, M., University of Maryland, College Park, MD; Poster, D.L.*, Neta, P., National Institute of Standards and Technology, Gaithersburg, MD. As part of an investigation on the use of ionizing radiation for the remediation of PCB contaminated material, the degradation of PCBs in oil induced by ionizing radiation was examined. A tetrachlorobiphenyl congener was solubilized in oil and subsequently irradiated by a ^{60}Co γ source. Rates and extent of dechlorination were determined, and intermediate species and by products were quantified. In addition, pulse radiolysis was investigated to measure rates of reduction of PCBs. Experiments were also conducted using a mixture of PCBs in oil. Experimental methodologies and results of this work will be presented. Applications of these methods to oils and the use of an oil standard reference material (SRM) that is well characterized for PCBs content will also be discussed.

PHA127 **Evaluation of the Efficacy of Photocatalysis combined with biofilm uptake for the Attenuation of Organic Contaminants.** Friesen, D. A., Headley, J.V.*, National Water Research Institute, Saskatoon, SK, Canada; Neu, T., UFZ-Magdeburg, Magdeburg, Germany; Langford, C. H., The University of Calgary, Calgary, AB, Canada. Photocatalysis by TiO_2 with UV or sunlight irradiation shows promise as a method for remediation of waters contaminated by organic compounds. Total degradation by this method consists of oxidation of the organic to CO_2; however, in some cases total destruction is not economically feasible. As well, intermediates recalcitrant to further oxidation occur in certain cases. Thus, there is interest in the combination of photocatalysis with biological treatment methods. We present studies of the combination of photocatalyzed degradation of N-methylpyrrolidinone (a solvent in the electronics industry) with biofilm uptake. Photocatalysis by TiO_2 leads to formation of a primary intermediate, N-methylsuccinimide. Biofilm uptake of this species is rapid, with over 80% uptake of the photogenerated intermediate over a 24 hour period. The results underline the promise of combined methods for remediation of contaminated waters.

PHA128 **Temperature-Dependence of the Hydrolysis of Endosulfan.** Sarunya Hengpraprom, C.M.Lee, A.W. Elzerman, and J.T. Coates, Environmental Engineering and Sciences, Clemson University, Anderson SC 29625-6510. Runoff of pesticides into estuaries may cause toxicological risk to aquatic organisms. Of particular concern is the runoff of the insecticide, endosulfan, which is commonly applied to tomato fields in many agricultural areas in the State of South Carolina. The objective of this study was to examine how temperature can enhance the hydrolysis of endosulfan in distilled water, river water, and seawater. Hydrolysis of endosulfan was studied over a temperature range of 30-40 °C and at pH 5 and 8 under sterile and non-sterile conditions. River water and seawater were collected from the Edisto River and Edisto Beach State Park in South Carolina. The disappearance of endosulfan and its metabolites were tracked for approximately four half-lives by gas chromatography using electron capture detector (GC-ECD). Pseudo first order kinetic will be observed for the disappearance of endosulfan under pH and temperature conditions. Calculated rate constants for the hydrolysis will be presented as well as the activation energy. Information from this study will be helpful for the better understanding of the kinetics of the hydrolysis of endosulfan, and useful for improving management practices to prevent exposure.

PHA129 **Halogenated By-products Formed Through Reactions of Chlorine with Tea Polyphenols.** Wu, W.W.*, University of Florida, Gainesville, FL; Chadik, P.A., University of Florida, Gainesville, FL; Davis, W.M., Environmental Laboratory, ES-P, Waterways Experiment Station, Vicksburg, MS; Powell, D.H., University of Florida, Gainesville, FL. Disinfection by-product (DBP) formation and chlorine consumption were investigated in the preparation of instant tea with water containing chlorine residual. Two instant tea samples formed as much DBPs as aquatic humic substances under the same reaction conditions (Temperature = 25 ± 2 °C, pH = 7.0 ± 0.1 and 8.5 ± 0.1, Br^- concentration = zero and 250 µg/L, Non-purgeable organic carbon (NPOC) = 5 mg/L, Chlorine dose = 12.5 mg/L, Chloramine dose = 6.25 mg/L as Cl_2, Contact time = 24 hours.), although the concentration of tea used for consumption typically is much higher than the concentration of humic substances in natural waters. At typical tea concentrations (NPOC ≈ 1000 mg/L), substantial levels of TOX (164-196 µg/L as chloride) were generated when chlorinated at 4 mg/L. Use of boiled water in preparation of instant tea is suggested if the residual free chlorine in the tap water is high.

PHA130 Potential Effects of Fire on the Spatial and Temporal Distribution of Mercury in Sediments of an Arid-lands Reservoir. Canavan, C.M. New Mexico State University, Las Cruces, NM; Caldwell, C.A. USGS/BRD, New Mexico Cooperative Fish and Wildlife Research Unit, Las Cruces, NM; Bloom, N. Frontier Geosciences Inc., Seattle, WA. Caballo Reservoir is located in southcentral New Mexico on the Rio Grande and is managed for flood control and irrigation. As a result, the reservoir undergoes seasonal volume fluctuations that inundate plants and soil. Spatial and temporal variability of total mercury (THg) and monomethlymercury (MMHg) were examined in sediments collected monthly from July 1995 to June 1996. Concentrations were greatest in sediment at a site located within a shallow area containing an extensive littoral zone (Palomas) compared to five sites in deep water containing minimal littoral zone. Concentrations of MMHg in sediments were greatest at the Palomas site (2.5-12.5 ng/g, dry weight) compared to MMHg in sediment from the five sites in deep water (< 1.0 ng/g). The ratio of MMHg to THg (an gross index of methylation activity) was greatest in sediment from the Palomas site (5.4-33.8%) compared to sediments from the five sites in deep water (0.0-3.6%). The effects of seasonal flooding on mercury concentrations in Palomas sediment was likely overshadowed by the result of two unrelated events. On 4 July 1995, a fire burned approximately 8,799 hectares near the headwaters of Palomas Creek which flows into Caballo Reservoir. The fire was followed by summer monsoon rains which carried an extensive amount of charred vegetative material down Palomas Creek dispersing the material at the Palomas site. The fire followed by rain may not only have brought increased organic matter into the reservoir, but may have mobilized and transported mercury from the watershed.

PHA131 Trifluoroacetic and other haloacetic acids (HAAs) in Canadian Freshwater and Marine Environments. Scott, B.F., Muir, D.C.G, Spencer, C., Struger, J. Environment Canada, Burlington, ON; Macdonald, R. and Mclauglin, F., Fisheries & Oceans, Sidney BC; Welch, H., Fisheries & Oceans, Winnipeg MB Canada. HAAs, especially mono- and trichloroacetic acid have been identified as an environmental concern in Europe over the past 10 years due to their phytotoxicity. Trifluoroacetic acid (TFA), a very stable degradation product of HFC and HCFC replacements for ozone-destroying chloro-fluorocarbons, has been predicted to increase 10x as a result of the growth in use of its precursors. We have determined TFA and other haloacetic acids in the surface waters of the Laurentian Great Lakes and four lakes in the other regions of Canada. Surface waters of the smaller lakes and Lake Superior have low concentrations of the TFA, (< 1 to 20 ng/L) whereas the other Great Lakes have values 7 times greater which range up to 150 ng/L. Other HAAs have concentrations ranges of <1 to 1000 ng/L. Storm water retention ponds situated in southern Ontario were found to contain TFA at the 400 ng/L level. Concentration values of other HAAs were determined to be at the ug/L level in these ponds. These ug/L values approach levels which may be phytotoxic to aquatic plants. To examine sources and retention of HAAs, depth profiles of TFA and other HAA concentrations were conducted each of the Canadian Great Lakes. These findings will be compared to a depth profile conducted in the Canadian Basin in the Arctic Ocean, in water masses of known residence time, to provide insight into whether there are significant natural sources of HAAs.

PHA132 Continuous and Synoptic Monitoring of Water Quality in a Contaminated Watershed to Characterize a Highly Dynamic Aquatic System for an Ecological Risk Assessment. Henningsen, G.*, U.S. EPA, Denver, CO; Hanley, J., U.S. EPA, Denver, CO; Edelman, P., U.S. Geological Survey, Pueblo, CO; Beatty, B., C.D.M., Denver, CO; Fischer, A., C.D.M., Denver, CO. The Alamosa River watershed in southern Colorado drains a rugged mountainous region as it flows east towards the Rio Grande River about 50 miles from the head-waters. This aquatic system is impacted by a major former mining operation and by unusually elevated levels of natural metals and hydronium ions (low pH). Snow-melt and storm events result in highly dynamic water quality and stream-flows, which complicate the exposure analyses in the ecological risk assessment. The US EPA Region VIII and the USGS, under review by a Superfund site BTAG (biological technical assistance group), designed and conducted a real-time continuous-monitoring sampling and analyses for pH, conductivity, and stream-flow at several Alamosa River segments for nearly 3 years. More thorough and periodic, about monthly, samplings and analyses were also conducted on the metallic contaminants of concern and general water quality. This analytical approach has proven effective in describing the nature and extent of contaminants plus water quality, and their variability within this watershed, for use in an ecological risk assessment. Results of these temporal measurements show the presence of four major flow regimes in five major spatially distinct stream segments. Both acute and chronic exposures were able to be characterized for use in comparing to risk-based benchmarks of aquatic toxicologic effects for various receptors of concern in the watershed. Repeated sampling permitted statistical analyses of the ranges of variability and uncertainty for the exposure analyses and effects analyses for aquatic receptors in the ecological risk assessment.

PHA133 Assessment of Polycyclic Aromatic Hydrocarbon (PAH) Exposure to *Ictalurus punctatus* in Two Urban Reservoirs using Semipermeable Membrane Devices and the Analysis of PAH Bile Metabolites. Moring, J.B, U.S.Geological Survey., Austin, TX; Burkhardt, M.R., U.S.Geological Survey, Denver, CO. Polycyclic aromatic hydrocarbons were detected in surficial bed sediments of two urban reservoirs in Dallas Texas at concentrations that exceeded State of Texas and Federal guidelines for the protection of aquatic life. Assessing exposure pathways of PAHs to aquatic biota is difficult because parent PAH compounds typically do not bioaccumulate, particularly in fish. Semipermeable membrane devices were deployed in the water column of the two reservoirs as surrogates for aquatic biota to determine the potential for the exposure and uptake of non-metabolized PAHs in the dissolved phase. Pyrene, chrysene and benzo(b)- and benzo(k)fluoranthene were the most common PAHs concentrated in the SPMDs, and concentrations in the SPMDs were higher in water overlying areas where PAH concentrations were highest in surficial sediments. Total PAH metabolites in the bile of *Ictalurus punctatus* (Channel Catfish) were measured as benzo(a)pyrene equivalents to assess the exposure of fish to PAHs in the two reservoirs. Concentrations of PAH metabolites ranged from 1.47 to 13.7 mg/L, and were generally higher in Channel Catfish from an urban reservoir dominated by non-industrial urban land uses in the watershed than from a reservoir with contamination dominated by discharges from adjacent industrial facilities.

PHA134 Variation of K_{oc} in Sediments from Narragansett Bay and Long Island Sound: Importance of Organic Carbon Quality. Burgess, R.M.*; Ryba, S.A.; Cantwell, M.G.; U.S. EPA NHEERL, Narragansett, RI; Gundersen, J.L. U.S. EPA NHEERL, RTP, NC. A discussion has been waging in the literature as to the importance of organic carbon quality to the partitioning of nonpolar organic contaminants to soils and sediments. Studies performed with pure organic matrices (e.g., cellulose, lignin) and those with actual soils and sediments often generate contradictory results as to the significance of organic carbon quality. The current study was conducted to investigate the importance of organic carbon quality on the partitioning of three nonpolar organic chemicals (Lindane, fluoranthene, 2,2',5,5' - tetrachlorobiphenyl) to five sediments from Narragansett Bay and Long Island Sound. Geochemical information, including elemental composition, stable carbon isotopes and grain size, show Narragansett Bay sediments are heterogeneous receiving organic carbon from at least two inputs: terrestrial/anthropogenic and planktonic sources. For the pesticide Lindane and the PCB, K_{oc}s were statistically different by site in only 5% of comparisons ($\alpha = 0.05$). Fluoranthene K_{oc}s were far more variable with 30% of comparisons detecting significant differences. Attempts to normalize equilibrium fluoranthene concentrations by other geochemical variables (e.g., polarity index) to reduce inter-site variability achieved various levels of success. Partitioning predictions by a two compartment model involving natural organic and soot carbon components corresponded closely to empirical data. This study indicates the major geochemical factor controlling the partitioning of Lindane and the PCB to marine sediments is organic carbon quantity; organic carbon quality may have some influence but the magnitude of this effect is relatively minor. Conversely, the partitioning of fluoranthene, and possibly other PAHs (as shown also by others), appears to be strongly affected by other sediment characteristics (e.g., soot).

PHA135 Effect of Industrial effluents on water and soil Qualities in Nnewi, Nigeria. 1. Orisakwe,O.E.* Nnamdi Azikiwe University Nnewi Campus Anambra state, Anambra State, Asomugha, R. Nnamdi Azikiwe, Awka Campus, Anambra State. Obi, E., Chilaka, K.C., Afonne, O.J.,Dioka,C.E., Nnamdi Azikiwe University Nnewi Campus. Nnewi is the commercial nerve centre and the most highly industrialised city of the east of the River Niger in Anambra State, Nigeria, West Africa. These industries specialise in the manufacturing of automobile spare parts and electrical accessories. We have investigated the impact of chemical arising from these industries on water and soil qualities in Nnewi, Nigeria. Higher levels of Iron, copper, lead and manganese were found in the deep than top soil. Soil samples from Ibeto industries Ltd had the highest levels of manganese (3617ppm) and 664ppm of lead while soil from RIMCO industries showed hihgest level of lead (746ppm). The pH of the soil ranged from 3.5 - 6.4. River Ele had highest level of lead (192.5mg/l) at the source than middle and lower courses which had 25mg/l each. The total hardness, salinity and biological oxygen demand of River Ele ranged from 14 - 35mg/l, 121.6 - 211.2mg/l and 1.9 - 2.7mg/l respectively from source to lower course. The highest level of volatile and non - volatile solid were 1.6 and 1.0 mg/l respectively. There was absence of live or dead macroorganism along the course of River Ele. Since there was elevated soil lead levels from most of the industrial sites, we recommend a control measure to reduce lead exposure to adjoining communities.

PHA136 Relationships Between LC50 and Body Burden of Copper in the Invertebrates *Hyalella azteca* and *Daphnia magna*. Collyard*, S.A., Boese, C.J., Bergman, H.L. and Meyer, J.S., University of Wyoming, Laramie, WY, USA. Acute toxicity of transition metals to aquatic biota is strongly influenced by various physical-chemical parameters that affect metal bioavailability. These parameters include pH, hardness, alkalinity, and dissolved organic carbon (DOC). Recent studies have demonstrated metal concentrations in aquatic biota are potentially more useful predictors of toxicity than are aqueous concentrations of metals. A biotic-ligand model that incorporates both metal speciation and competitive binding of cations to binding sites on the gill surface successfully accounts for high variability in toxicity of total dissolved Cu to fish as water hardness and pH are varied. However, the biotic-ligand model has not yet been tested with aquatic invertebrates. We demonstrate that sigmoidal dose-response curves result when percent mortality is plotted versus Cu body burden in *Daphnia magna*, *Hyalella azteca* and *Chrironomus riparius*, with shapes similar to the dose-response curves when percent mortality is plotted versus total dissolved Cu concentration. Yet the Cu body burden at 50% mortality in *D. magna* remains constant across a range of pH and water hardness, whereas the total dissolved Cu LC50 varies considerably. Similar results will be shown for *H. azteca*, for which we also will show that stability constants for Cu accumulation can be estimated. Thus, the biotic-ligand model can be used to predict acute toxicity of Cu to aquatic invertebrates as well as to fish across a range of water quality conditions.

PHA137 Cu Uptake Kinetics and Critical Gill Cu Concentrations in Chinook Salmon Fry. Lipton, J., Welsh, P.G., Hagler Bailly Services, Boulder, CO; Playle, R., Wilfrid Laurier University, Waterloo, Ontario, Canada. One approach to evaluating metal bioavailability/toxicity to fish is to treat the gill as an organic ligand and predict metal accumulation on the gill. Gill residues then are presumed to be related to acute lethality at some critical toxicant concentration. We performed an experiment to evaluate Cu uptake and gill Cu residues in survivors vs. moribund fish. We exposed chinook salmon (*Oncorhynchus tshawytscha*) fry for 48 hours to 100 μg/L Cu in soft water. Ten fish were sampled at test initiation and after 1, 2, 4, 8, 24, and 48 hours. In addition, we sampled aquaria every 30 minutes (from 8-48 h) to remove dead/moribund fish with minimal interference from post-mortality Cu accumulation. Gills were immediately excised and gill-Cu concentrations determined by GFAAS. Cu concentrations on the fish gill increased dramatically over the first 4 hours of Cu exposure from 28 to 266 nmol Cu/g dry wt. Gill-Cu concentrations subsequently decreased to 100 nmol Cu/g dry wt, presumably due to activation of compensatory mechanisms. This pseudo-equilibrium between the fish gill and the water occurred after 24 h Cu exposure. Dead fish had gill-Cu concentrations similar to live fish after 24 and 48 h Cu exposure. We discuss uptake kinetics and whether these data support the critical residue concept.

PHA138 Modeling Chronic Thresholds For Toxicity-physiological effects of chronic Cu exposure to rainbow trout. Taylor, L.N.*, McGeer, J.C., Wood, C.M. and McDonald, D.G. McMaster University, Hamilton, Ontario. Impairment of growth is one of the most commonly used endpoints for the evaluation of the chronic toxicity of environmental contaminants and for the setting of chronic no-effect thresholds. In this study, we test the hypothesis that growth reduction is not necessarily the most sensitive indicator of chronic copper toxicity. Juvenile rainbow trout (*Oncorhynchus mykiss*) were exposed to sublethal waterborne copper for 30 days in moderately hard and soft water to evaluate six endpoints: tissue residues, acclimation (i.e. increased lethal tolerance), whole body electrolytes, swim performance, gill copper binding characteristics and growth. Copper exposure had no effect on growth and swimming performance but did result in elevated tissue metal burdens, decreased whole body electrolytes and differences in gill-metal permeability. Our results suggest that growth can be one of the least sensitive indicators of copper toxicity depending on previous exposure concentrations, the level of chronic adaptation to the metal that was achieved and differences among feeding regimes.

PHA139 Why Is the Slope of the ln(LC50) vs. ln(Hardness) Adjustment Equation for Copper Approximately Equal to 1? Meyer, J.S., University of Wyoming, Laramie, WY, USA. In the U.S. Environmental Protection Agency's water quality criteria for Cu, the adjustment factor that accounts for the protective effect of water hardness is based on a regression of ln(LC50) vs. ln(hardness) whose slope is 0.9422. This regression slope can be explained by deriving a general equation for metal binding to a biotic ligand, an equation in which (1) aqueous speciation of Cu and (2) competitive binding of Cu^{2+}, Ca^{2+}, and H^{+} to the biotic ligand are incorporated and the amount of Cu bound to the biotic ligand at 50% mortality is assumed constant. However, the slope of approximately 1 only applies when hardness and alkalinity are co-varied across a range of intermediate hardness and alkalinity. As hardness approaches zero, the regression slope approaches zero; and as hardness becomes extremely high, the slope depends on the rate of change of alkalinity. If alkalinity is constant as hardness is varied at extremely high hardnesses, the regression slope approaches 1. If alkalinity co-varies with hardness at extremely high hardnesses, the regression slope approaches 2. Thus, the hardness-adjustment equation for Cu can be derived from a mechanistic model instead of being just an *ad hoc*, ln-transformed linear regression equation. Additionally, the regression slope should not be assumed constant. A biotic-ligand model that accounts for hardness and alkalinity (and pH, dissolved organic carbon, and suspended solids) in the exposure water can provide a more effective way of predicting Cu LC50s in waters in which complex, nonlinear interactions among water-quality variables occur.

PHA140 The Effect of Hardness, pH, and TOC on Copper Toxicity. Rasnake, W.J.*; Dietz, K.; Wagner, M.B.; Gruber, D.; Biological Monitoring, Inc., Blacksburg, VA. The U.S. EPA water quality criterion for copper is based exclusively on water hardness values. Concerns with the conservative nature of the criteria values, as presently calculated, have spawned numerous site-specific criteria/standard modification studies. Water effect ratio studies for copper, conducted by this firm, have consistently yielded an increase in the water quality standard by more than one order of magnitude. BMI has conducted several laboratory-based copper toxicity studies. Static acute bioassays have been conducted on *Ceriodaphnia dubia* at two different hardness values, two different pH values and at two different TOC values. Of all the variables, TOC overwhelmingly had the greatest effect on Copper toxicity.

PHA141 The Effect of Different Ligands on the Bioavailability of Dietary Copper in Rainbow Trout. Clearwater, S.J.*; Baskin, S.J.; Wood, C.M.; and McDonald, D.G. McMaster University, Hamilton, Ontario, Canada. Mammalian research suggests that the major factor influencing the bioavailability of dietary Cu is diet composition. For example, Cu bioavailability can be reduced by a competing ligand (e.g. zinc) or by complexation to a non-soluble dietary constituent (e.g. fibre), or increased by complexation to a nutrient (e.g. amino acid). We have developed an *in vivo* technique to dose juvenile rainbow trout (200-250 g) via a surgically implanted stomach catheter with 2.4 μmol of radio-labelled Cu in the presence or absence of ligands. Using this approach, Cu was partitioned into fractions bound in or on the gut tissue, absorbed internally (the bioavailable fraction), remaining in the gut lumen, and excreted. The different treatments were Cu with glucose (i.e. no ligand, control), with Ca and with Zn (competing ligands), nitrilotriacetic acid (NTA, complexing agent) and cysteine (CYS, source of SH groups). By 48h after infusion only 5% of the Cu dose (minus ligand) had been absorbed. The largest fraction of this was found in the liver. NTA and Ca increased bioavailability of the dose (to 9.1% and and 7.5%, respectively) while Zn and cysteine reduced bioavailability (to 3.7% and 3.1%, respectively). Although NTA and Ca had similar effects on absorption through the gut, with NTA there was less copper bound to the gut tissue than with Ca. Moreover, with NTA, Cu was distributed primarily to liver, kidney and plasma, whereas with Ca, Cu was distributed mainly to the liver and bile. The reduced bioavailability of Cu+CYS was unexpected and may be because the complex was insoluble in the digestive tract. These data allow us to explore the different pathways by which dietary copper uptake occurs and to understand the important factors that influence copper bioavailability.

PHA143 Development of a Site-Specific Water Quality Benchmark for Hexavalent Chromium. Duda, D.J.*, Barber, T.R., Fuchsman, P.C., Sferra, J.C., McLaren/Hart, Cleveland, OH. Hardness has been shown to lower the bioavailability, and hence toxicity, of various metal ions. While ambient water quality criteria for trivalent chromium are calculated as a function of hardness, the criteria for hexavalent chromium (11 μg/L chronic and 16 μg/L acute) were established without regard to hardness due to a lack of appropriate data (USEPA, 1984). We reviewed more recent studies to determine whether toxicity due to hexavalent chromium can be expressed as a function of hardness. While the toxicity of hexavalent chromium to fish (which are relatively insensitive) shows no relationship with hardness, several publications indicate that toxicity of hexavalent chromium to cladoceran species (which are most sensitive) is affected by hardness. The water quality criterion for hexavalent chromium is based on the most conservative studies available, which were conducted using cladoceran species exposed in soft water (e.g., 45 mg/L $CaCO_3$). Differences among Site-specific water quality conditions limit the applicability of this criterion. We calculated Tier II water quality benchmarks applicable to waters with moderate (51-300 mg/L $CaCO_3$) and very high (>300 mg/L $CaCO_3$) hardness levels. These benchmark were approximately 3 and 10 times higher than the chronic water quality criteria for moderate and high hardness, respectively. These benchmarks were used to assess potential impacts of hexavalent chromium on biological communities in a Northeast Ohio river characterized by hard water conditions and bordered by a former chromium product operations plant. Measurements of Biological community quality appeared to correspond to physical habitat conditions rather than chromium concentrations, as predicted by the site-specific benchmark for hexavalent chromium.

PHA144 The influence of organic matter on the bioavailability and toxicity of cadmium and copper in metal-spiked sediments. Besser, J.M.*, Ingersoll, C.G., Brumbaugh, W.G., and May, T.W. USGS Biological Resources Division, Columbia MO. Proposed models for sediment quality criteria for cationic heavy metals predict the absence of metal toxicity in sediments which contain "excess" acid-volatile sulfide (AVS), but do not consider the influence of other sediment constituents which may reduce metal bioavailability. Organic matter may reduce the bioavailability of metals in oxic sediments, or in sediments where metal concentrations exceed AVS concentrations. Simple models of metal adsorption by sediment organic matter could be used to improve models for predicting the bioavailability of metals in sediment. However, the capacity of sediment organic matter to adsorb metals may vary, depending on the types and concentrations of metals and organic matter present. The objectives of this study were: (1) to compare the ability of different types of sediment organic matter to adsorb metals and reduce metal toxicity; and (2) to evaluate whether the influence on sediment organic matter on metal toxicity can be predicted from simple adsorption studies. Interactions of two metals (cadmium and copper) and two types of organic matter (cellulose and humus) were evaluated in batch adsorption studies and in toxicity tests with metal-spiked formulated sediments. Short-term (24- to 48-hr) batch experiments, consisting of a series of metal concentrations and varying adsorbent:solution ratios, were used to construct adsorption isotherms. Toxicity tests with the amphipod, *Hyalella azteca*, were conducted with metal-spiked formulated sediments containing differing types and concentrations of organic matter. Concentrations of metals in sediment porewater and toxic effects in *H. azteca* were compared to predictions based on toxicity curves for aqueous metals and metal adsorption isotherms for different types of organic matter.

PHA145 Modelling Cu and Cd Uptake in Rainbow Trout, *Oncorhynchus mykiss*, Following Chronic Sublethal Exposure to Cu or Cd. McGeer, J.C.*, Hollis, L. Alsop, D. Taylor, L. McDonald, D.G. and Wood, C.M.; McMaster University, Hamilton, Canada. The chronic toxicity of waterborne heavy metals is poorly understood and can be complicated by the phenomenon of acclimation, an enhanced resistance to subsequent challenges. Our studies have examined how acclimation to heavy metals alters the correlation between components within existing acute toxicity models. Juvenile rainbow trout were exposed to either 3 µg/L Cd or, 75 µg/L Cu for one month in hard water (120 mg/L as $CaCO_3$) with minimal mortality and no effect on growth. Chronic sublethal exposure to metal resulted in acclimation and significant accumulations of metal (either Cd or Cu) in the gill, kidney and liver. Accumulations of Cd were highest in the gill while those of Cu were greatest in the liver. Following a month of exposure to either Cd or Cu, metal uptake into gills, blood, liver, kidney, gastrointestinal tract and whole body was studied over time using radioisotopes (^{109}Cd or ^{64}Cu). Acclimation to Cd resulted in a reduction in the rate of Cd uptake into the gills and whole body compared to uptake rates of naive fish exposed to 3 µg/L Cd. When challenged at the higher level of 10 µg/L Cd, acclimated fish had significantly higher rates of gill Cd uptake and previous exposure to Cu similarly enhanced Cd uptake. In addition, chronic metal exposure resulted in altered routing of new metal through different body compartments. These results illustrate the changes in metal uptake and internal distribution that occur during chronic sublethal exposures. Our goal is to model metal uptake and internal distribution and to predict both acute and chronic toxicity in acclimated fish. Supported by NSERC through a Stragetic Research Grant and a PDF and by Cominco Ltd., Falconbridge Ltd., and the International Copper Association.

PHA146 Untested Assumptions in Water Effect Ratio Testing. Welsh, P., Lipton, J., Hagler Bailly Services, Inc., Boulder, CO., and Chapman, G., Paladin Water Quality Consulting, Corvallis, OR. One approach outlined by the U.S. EPA for derivation of site-specific aquatic life criteria involves the development of water effect ratios (WER). This approach entails multiplying the national ambient water quality criterion value by an experimentally derived WER, where the WER is defined as the ratio of the toxicity of a metal in site water to the toxicity of the same metal in standard laboratory water. The laboratory water is typically formulated to match the total hardness of the site water and differences in toxicity are assumed to be due to other modifying constituents in the site water (e.g., DOC). The current WER methodology may result in site specific water quality criteria that is underprotective. We will present data to address a number of untested assumptions or omissions in the current U.S. EPA WER guidance document that address this, including the importance of matching the ionic composition of the test water (e.g., Ca:Mg ratios vs. total hardness), the importance of fish acclimation to test water hardness, the influence of multiple metals on WERs, and the testing methodology used (static renewal vs. flowthrough). Overall, we feel that these issues have the potential to result in WERs that are biased high (i.e., underprotective) or represent a source of uncertainty that needs to be addressed.

PHA147 Metals Toxicity to Aquatic Species Associated with Tribal Wetlands, Swamp Creek, Wisconsin. Castle, C.J.*, Colorado State University, Ft. Collins, CO; Nimmo, D.R., Midcontinent Ecological Science Center, BRD/USGS, Ft. Collins, CO; Preul, M.R., Colorado State University, Ft. Collins, CO. This is a progress report on baseline conditions and the toxicity of metals to various species in Swamp Creek and Rice Lake, Wisconsin. Baseline tests were conducted on surface and pore waters derived from sediments in the vicinity of the Nicolet Mine near Crandon, Wisconsin. Test species were *Ceriodaphnia dubia, Pimephales promelas, Hyalella azteca,* and *barnyard grass, Echinochloa crusgalli.* Next, water from Swamp Creek was fortified (spiked) with copper to determine the metals's toxicity to resident species in natural water. Results of the copper-spiked tests have raised issues regarding the sensitivity of culturally-valued species such as walleye, *Stizostedion vitreum*, and wild rice, *Zizania aquatica* to copper if the mine is developed.

PHA148 Derivation of Ambient Water Quality Criteria for MTBE: Toxicity to Selected Freshwater Organisms. K.P. Christensen*, J.R. Hockett, W.A. Stubblefield, ENSR Environmental Toxicology, Fort Collins, CO; A. Steen, American Petroleum Institute, Washington, DC; J. Grindstaff, Columbia Analytical Services, Inc, Kelso, WA; D.C.L. Wong, Shell Oil Products Co, Houston, TX; and W.R. Arnold, Exxon Biomedical Sciences, Inc., East Millstone, NJ. MTBE is a water-soluble fuel oxygenate whose production and use has raised concerns regarding its environmental effects. To address those concerns and establish safe concentrations in surface waters, existing aquatic toxicity data were evaluated, and new freshwater toxicity tests were conducted to satisfy EPA's database requirements for deriving ambient water quality criteria. Existing studies meeting EPA's requirements addressed acute toxicity of MTBE to three families o freshwater organisms: Cladocera (*Daphnia magna* and *Ceriodaphnia dubia*), Cyprinidae (fathead minnows, *Pimephales promelas*), and Salmonidae (rainbow trout, *Oncorhynchus mykiss*). Available chronic data did not meet EPA's requirements for inclusion in the AWQC database. In a collaboration between an industry group, EPA, and contract laboratories, additional testing was undertaken to meet the database requirements for acute endpoints (EC_{50}s) for five additional families of freshwater animals, including Hexageniidae (mayfly nymph, *Hexagenia* sp.) Chironomidae (midge larvae, *Chironomis* sp.), Talitridae (amphipod, *Hyalella* sp.), Physidae (snail, *Physa* sp.) and Centrarchidae (bluegill sunfish, *Lepomis macrochirus*). Chronic tests were undertaken for two freshwater species, *Daphnia magna* and fathead minnows, and a short-term chronic test conducted with the freshwater alga, *Selenastrum capricornutum*. The results of the acute toxicity tests show that, in general, MTBE exhibits low toxicity to freshwater species, with EC_{50}s in the hundreds of milligrams per liter. The data objectives for meeting EPA's requirements for inclusion in an AWQC database will be discussed along with the endpoints observed and the technical aspects of conducting toxicity tests with a volatile, solvent test substance.

PHA149 MTBE Water Quality Criteria Database Development: a Collaborative Effort. Mancini, E.R.*, ARCO, CA; Arnold, W.R., EXXON, NJ; BenKinney, M.T., Mobil, NJ; Gostomski, F.E., USEPA, DC; Rausina, G.A., Chevron, CA; Steen, A., API, DC; Wong, D.C.L., Shell Development Company, TX. The alkyl ether, methyl tertiary-butyl ether (MTBE), is widely used in modern gasolines to achieve various fuel formulation and emissions objectives. The detection of MTBE in some groundwater and surface waters has raised concerns about potential effects on aquatic organisms. Federal ambient water quality criteria for the protection of aquatic life do not currently exist for MTBE. In response to these concerns, a collaborative effort was undertaken to complete freshwater and marine aquatic toxicity databases, as required by USEPA guidance, for deriving numerical criteria. Prior to project initiation, existing acute exposure freshwater data for six species ranged from 195 to 2500 mg/l while chronic exposure, NOEC data for two species were 203 and 234 mg/l. Acute exposure toxicity data for five marine species ranged from 44 to >2500 mg/l. This collaborative effort included the American Petroleum Institute (API), the Oxygenated Fuels Association (OFA), the Western States Petroleum Association (WSPA), several member companies of these organizations and technical representatives from the USEPA who participated in oversight, advisory and critical review roles. Two commercial aquatic toxicology testing laboratories were competitively selected to conduct freshwater and marine testing programs. Sixteen additional acute and chronic exposure data for eight freshwater and seven marine species were developed under GLP-consistent protocols and procedures in order to complete the criteria databases. Testing was conducted under the oversight of industry study monitors and independent, third-party auditors. Based upon the completed aquatic toxicity databases and consistent with the existing guidance, freshwater and marine, acute and chronic concentrations for the protection of aquatic life were calculated and are presented.

PHA150 Evaluation of the applicability of risk assessment methodologies for essential elements. Karman, C.C.*; Jak, R.G., TNO Dept. for Ecological Risk Studies, Den Helder, The Netherlands. Most of the methods developed for ecological risk assessment are based upon the assumption that zero environmental concentrations will offer full protection for the ecosystem. However, chemicals exist that are essential for life and should be available for uptake in such concentrations that an optimal internal concentration can be maintained. For these, so-called, essential elements, regulation mechanisms have evolved to maintain a desirable internal level at which optimal functioning of an organism can be achieved. Therefore, the naturally variability in bioavailable natural background concentration ranges, which vary on both a spatial and a temporal scale, and the capabilities of organisms to condition to ranges of background levels should be considered in risk assessment studies. The existing methods for ecological risk assessment evaluated in this study are using an "assessment factor approach", a "statistical extrapolation approach" and a "homeostasis approach". Although these methods may have been developed to serve different objectives, they usually are applied intermixed to calculate environmental threshold levels. To facilitate the evaluation, 6 evaluation criteria have been defined. Four of these criteria are considering the way in which methods account for factors related to biota (species representativeness, inter-species variation, laboratory-field extrapolation, deficiency-toxicity). Two other criteria consider factors affecting the exposure of organisms to a chemical (bioavailability, background concentration). The evaluation lead to the conclusion that mainly the data-requirements determine whether risk assessment methods are applicable for essential elements. These data-requirements are best defined for the "homeostasis approach", since it has been developed for essential elements. When, however, these data requirements are applied to the other methods, these comply to the criteria as well.

PHA151 How dirty is that stormwater detention pond in your neighbourhood and who lives in it? Environment Canada investigates the potential risk of contaminants in constructed wetlands to wildlife. Bishop, C.A.*, Struger, J., Dunn, L. Canadian Wildlife Service and Ecosystem Health Division, Environment Canada, Burlington, ON, and Bedard, D., Standards Development Branch, Ontario Ministry of the Environment, Etobicoke, ON, Canada. We assessed wildlife use in 15 constructed stormwater ponds in Toronto and Guelph by monitoring each site for presence of amphibian, bird, reptile and fish species between April and October in 1997. Surface water samples were collected periodically throughout the study and were analyzed for physical characteristics, nutrients, metals, and major ions. Sediment samples were collected once from each site and analyzed for PAHs, PCBs, OC insecticides, metals, nutrients, TOC, and oil and grease. We also collected red-winged blackbird eggs and fish for contaminant analysis. Sediment samples were also collected and used to conduct bioassays with invertebrates and fish. In total, among all of these ponds, we observed 71 species of birds feeding or nesting; seven species of amphibians spawning at the sites; four species of reptiles at or adjacent to the ponds and six species of fish. Red-winged blackbird eggs from two locations in Toronto had low concentrations of PCBs and organochlorine pesticides. The concentrations were 0.26-1.13 ug/g pp'DDE and <0.67 ug/g total PCBs. Maximum concentrations of some water quality parameters were 3.96 mg/L of NO3-NO2, 0.722 mg/L of NH3, 6.31 mg/L of TKN, 0.72 mg/L of TP, 73.4 ug/L of Zn, 16.0 ug/L of Cu , and 17.5 ug/L of Pb. Maximum concentrations of some sediment quality parameters were 789 ng/g of total PCBs, 85 ug/g of PAHs, 535 ug/g of Zn, 52.4 ug/g of Cu, and 67.6 ug/g of Pb. These data will be discussed in relation to water and sediment quality guidelines, results of sediment bioassays, and possible effects on biota utilizing these facilities.

PHA152 Development and Construction of a Freshwater Treatment Wetland and an Intertidal Wetland Adjacent to the Houston Ship Channel. Quast, W.D.*[1]; Chang, C.C.[2]; Murray, J.L.[3]; Davidson, R.L.[1]; Kovski, J.R.[2], Williams, G.[2]; [1] Parametrix, Inc.[2] Radian International LLC[3] Mobil Mining and Minerals Company. In the past, extensive shoreline modification and poor water quality in the Houston Ship Channel (HSC) discouraged utilization of the waterway by aquatic biota. While water quality in the HSC has improved and some species of finfish and shellfish have returned to the system, habitat suitable for the growth and development of estuarine biota (intertidal wetlands) is scarce. The purpose of this project was to construct a system of wetlands that would increase the availability of high quality intertidal habitat adjacent to the waterway, and improve the quality of a treated effluent stream that is discharged to the waterway. A 7 hectare wetland with three hydraulic connections to the HSC was constructed to provide intertidal habitat for fish, shellfish, wading birds and other wildlife. The constructed wetland consists of intertidal marsh, high marsh, and transitional upland. At a higher elevation on the same site, a 6.9 hectare freshwater treatment marsh was constructed. The treatment marsh consists of a series of deep pools and shallow planting shelves along a meandering water-course. The marsh was primarily designed to reduce ammonia levels in a treated effluent stream. Shallow grassbeds and open water pools in the marsh will also provide high quality habitat for birds and terrestrial wildlife. When fully developed, this wetland system will provide habitat and sustenance for wildlife in the HSC, where high quality habitat is rare, and will further improve the quality of an effluent stream discharged to the waterway.

PHA153 **Risk Evaluation of Metals in a Pilot Wastewater Treatment Wetland.** McBrien, M.A.; Weier, J.A.*, Metcalf & Eddy, Inc., Wakefield, MA. Risks associated with metals in wastewater, sediment and aquatic vegetation were evaluated for two pilot systems designed to test the ability of wetlands to treat primary and secondary effluent produced at two wastewater treatment plants in Alexandria, Egypt. Constructed wetlands were planted with common reed (*Phragmites communis*), cattail (*Typha* sp.), bulrush (*Scirpus* sp.), water hyacinth (*Eichhornia crassipes*), and duckweed (*Lemna* sp.). During each phase of the six month wetlands pilot operation, samples of wastewater, sediment and aquatic vegetation were analyzed for priority pollutant metals. The constructed wetlands successfully treated both primary and secondary effluent to within limits dictated by Government of Egypt Law 48 for heavy metals and other water quality parameters. At the East Treatment Plant, the constructed wetlands reduced concentrations of copper, lead and selenium in influent to below USEPA chronic ambient water quality criteria, but not mercury. The constructed wetland at the West Treatment Plant also reduced the concentrations of copper and lead to below chronic ambient water quality criteria. In sediment, metals concentrations in both pilot systems decreased during operation with secondary effluent and increased at the completion of operations with primary effluent. The concentrations of most metals exceeded conservative ecological benchmarks for sediment when the pilot project was completed, but were generally less than benchmarks indicative of severe impairment. Concentrations of metals in plant tissue generally did not increase over the six month pilot operation. An assessment of metals in vegetation indicated that harvested aquatic plants could safely be fed to livestock as a portion of their diet.

PHA154 **Constructed Wetlands to Mitigate Agricultural Pesticide Runoff.** Moore, M.T.*, University of Mississippi, University, MS; Gillespie, W.B., Jr., ENTRIX, Houston, TX; Rodgers, J.H., Jr., TIWET, Clemson University, Pendleton, SC; Cooper, C.M. and Smith, S., USDA-ARS National Sedimentation Laboratory, Oxford, MS. Four constructed wetlands (59-73 m x 14 m) were amended with the insecticide chlorpyrifos as simulated cropland runoff and rainfall event equal to three volume additions to each wetland. Targeted aqueous concentrations of chlorpyrifos following rainfall simulation and wetland dispersion ranged from 0 µg/L (untreated control) to 733 µg/L. Water, sediment, and plant samples were collected weekly for 84 days from sites distributed longitudinally within each wetland and analyzed for chlorpyrifos using gas chromatography. Chlorpyrifos rapidly sorbed to sediment and plant material, and the half-life in water, in this experiment, was approximately 4-6 days. Approximately 58% of the chlorpyrifos was retained in the first 10-15 m of the wetlands. Sediments retained 25-72% of the chlorpyrifos, while 12-58% of the chlorpyrifos was retained by the plants (dominated by *Juncus* spp.). According to these mesocosm-scale results, downstream or receiving system impacts could be mitigated by using constructed wetlands as "buffer strips" for agricultural pesticide runoff. This research provides design parameters and fundamental answers concerning constructed wetland capabilities that are necessary for constructing field-scale systems within agricultural watersheds.

PHA155 **Effectiveness of Pilot-Scale Biological Treatment of Acid Mine Drainage in Cornwall, England.** Isobel Austin and Jim Wright, Environment Agency, Exeter, UK, Mike Cambridge and Quentin Hamilton*, Knight Piésold, Ashford, UK, and Phillip M. Rury, Arthur D. Little, Inc., Cambridge, Massachusetts, USA. Historical flows of acid mine drainage (AMD) from the abandoned Wheal Jane tin mine have exceeded four million gallons per day. The AMD is characterised by its low pH and complex chemistry. Since 1975, Knight Piésold has conducted AMD assessments, advised on the management of the AMD problem and developed potential remedial solutions for the Environment Agency. In 1992, Arthur D. Little evaluated AMD biological treatment technologies then employed in the USA on behalf of the Environment Agency and recommended a conceptual scheme for an experimental, pilot-scale AMD bioremediation system at the minesite. Independently in 1994, a pilot passive treatment plant consisting of three separate treatment trains designed by Knight Piésold was constructed at the site to treat approximately 2 l/s and has since been operated by Knight Piésold for the Environment Agency. Due to the complex chemistry and low pH of the minewater, a variety of passive treatment options was investigated. These included aerobic reed beds with and without pre-treatment (anoxic limestone drain or lime dosing), anaerobic cells, and rock filters. Typically, the median influent minewater has a pH of 1, with median total concentrations of iron 140 mg/l, arsenic 2.5 mg/l, copper 0.4 mg/l, cadmium 0.15 mg/l, zinc 80 mg/l, aluminum 50 mg/l and manganese 23 mg/l. Median iron removal ranged from 63% to 71% with arsenic being removed to below detection limits. Anaerobically, copper and cadmium were removed to below detection limits, with median zinc removal ranging from 45% to 86%. Manganese removal in the rock filters, although variable, has been as high as 97%.

PHA156 **Abatement of Ammonia and Biochemical Oxygen Demand in a Petroleum Refinery Effluent Using Constructed Wetlands.** Huddleston, G.M. III*, Clemson University, Pendleton, SC; Gillespie, W.B., Jr., University of Mississippi, Oxford, MS; Rodgers, J.H., Jr., Clemson University, Pendleton, SC. The purpose of this study was to evaluate the feasibility of constructed wetlands for tertiary treatment of a petroleum refinery effluent. Specific objectives were to decrease concentrations of ammonia-nitrogen (NH_3-N) and 5-day biochemical oxygen demand (BOD_5) and to reduce toxicity. Bench-scale wetlands were integratively designed and constructed in a greenhouse to simulate tertiary treatment systems for refinery effluent. These wetlands consisted of *Typha latifolia* planted in low-organic (0.2%), sandy sediment, and had 48-h nominal hydraulic retention times. To determine treatment efficiency, targeted constituents and aqueous toxicity were monitored in wetland inflow and outflow for three months. Following a 2-3 week stabilization period, NH_3-N and BOD_5 concentrations in refinery effluent were consistently decreased by an average of 95% and 80%, respectively. Survival of *Pimephales promelas* and *Ceriodaphnia dubia* (7-day, static renewal exposures) increased by more than 50% and 20%, respectively. Lowest- observed-effects concentrations based upon *C. dubia* reproduction were 12.5% wetland inflow and 100% wetland outflow. This study demonstrated the potential for constructed wetlands to decrease NH_3-N, BOD_5, and toxicity in this refinery effluent, and provided baseline information for full-scale wetland design.

PHA157 **Avian Reproductive Success and Selenium Bioaccumulation in a Constructed Wetland Receiving Treated Refinery Effluent.** Ohlendorf, H.M.*, Byron, E.R., Castleberry, M.A., Santolo, G.M, CH2M HILL, Sacramento, CA; Gala, W.R.*, Chevron Research and Technology Co., Richmond, CA; Hegedus, A.S. III, Chevron Products Company, Richmond, CA. The Chevron Richmond, CA refinery developed a 36-hectare constructed wetland for tertiary treatment of some of its wastewater effluent. Studies were conducted in 1994 and 1995 to determine the numbers of aquatic birds (waterfowl and shorebirds) using the constructed wetland, their reproductive success, and selenium bioaccumulation in food-chain organisms and bird eggs. Nest success was greater in the constructed wetland than typically observed for these species in reference wetlands. Selenium bioaccumulation was higher in food-chain organisms than predicted on the basis of relationships found for agricultural drainwater systems. Based on the results of the 1994-1995 studies, a 5-year management program was developed jointly with regulators and resource agencies to encourage bird use of the lower-selenium (downstream) portions of the constructed wetland, while discouraging bird use in the inflow (upstream) portion, where selenium concentrations are higher. The first year of monitoring results (1997) showed favorable results for the management program. Shorebird foraging and nesting have been eliminated in the upstream portion of the constructed wetland, and the selenium concentrations in food-chain biota and bird eggs in downstream portions were reduced by about 50 percent, when compared to similar samples from 1995.

PHA158 Toxicity, Uptake and Distribution of Simazine in *Myriophyllum aquaticum*: Implications for a Phytoremedial System. Wilson, P.C.*, Whitwell, T., and Klaine, S.J., Clemson University Clemson, S.C. This research focuses on the potential use of *Myriophyllum aquaticum* for removing simazine from contaminated water. Because of simazine's herbicidal activity, it is important for levels in solution not to exceed plant tolerance levels. Tolerance levels for *Myriophyllum aquaticum* were determined by dosing plants for 7 d with 0, 0.01, 0.03, 0.1, 0.3, 1.0, or 3.0 mg-simazine/L- nutrient media. Measurements of 7-d fresh mass production and lengthwise growth were taken. Simazine uptake and distribution within the plant was determined by dosing plants with 2.03 µCi ^{14}C-simazine (0.243 mg/L) for 1, 3, 5, or 7 d. Plant tissues were analyzed by combustion and liquid scintillation counting. Fresh mass production was reduced 47, 97, and 105% for plants exposed to 0.3, 1.0 and 3.0 mg/L, respectively. Likewise, lengthwise growth was reduced 18, 60, and 53% at the same respective concentrations. Plant uptake of simazine accounted for 8, 17, 30, and 49 percent of the original simazine in the dosing solution. This simazine was distributed primarily in the stems and leaves.

PHA159 Impact of Contaminated Litter from a TCE Phytoremediation Project and of major metabolites of TCE on Terrestrial Isopods and Aquatic Amphipods. Sorbet, M.M.*, TIWET, Clemson University, Pendleton, SC; Hooper, M.J., Texas Tech University, Lubbock, TX; Gordon, M.P., University of Washington, Seattle, WA; Newman, L.A., University of Washington, Seattle, WA; La Point, T.W., Texas Tech Univesity, Lubbock, TX. Phytoremediation of TCE in groundwater is a groundbreaking area of bioremediation research. However, the TCE metabolites trichloroacetic and dichloroacetic acid (TCA and DCA) and trichloroethanol (TCEtOH) have been detected in the tissues of poplars used for remediation. We are exploring the impact of contaminated poplar leaves on terrestrial isopods (*Armadillidium* spp.), which are primary leaf litter decomposers. We are also examining the effect of the detected TCE metabolites on *H. azteca* (important leaf decomposers in aquatic systems) and on two dominant microbes found on control leaves. Preliminary results from a 10-week juvenile isopod growth and survivorship experiment do not reveal significant differences between groups fed control vs. contaminated leaves. We likewise found no feeding preference for control vs. contaminated leaves in mature isopods, as measured by relative leaf masses consumed over time. Static 48-hour LC50 concentrations of 213, 217, and 365 mg/L were obtained for TCA, DCA, and TCEtOH, respectively, using juvenile *H. azteca* (as a follow-up study, TCA will be added to *H. azteca* cultures at sublethal lethals and the animals' growth, survivorship and reproduction rates will be monitored over a 10- to 12-week period). Trials have not shown that TCA, DCA or TCEtOH possess anti-microbial properties at levels detected in contaminated leaves. Overall, our findings suggest that the levels of contamination found in leaf tissue of poplars used to phytoremediate TCE are sufficiently low as to pose minimal risk to local natural populations of isopods, amphipods, and leaf-colonizing microbes. Funded by NIH ES04696.

PHA160 Pseudomonad inoculant enhancement of phytoremediation by Dahurian wild rye. Siciliano, S.D.* and Germida, J.J. University of Saskatchewan, Saskatoon, SK, Canada. Phytoremediation can be used to clean up contaminated soil sites, but in some cases the mechanisms of phytoremediation are unknown. In this study we investigated the mechanism by which bacterial seed of Dahurian wild rye (*Elymus dauricus*) increase 2-chlorobenzoic acid (2CBA) degradation in soil. Soil was amended with 450 mg kg^{-1} 2-chlorobenzoic acid (2CBA) and aged for 2 years, at which time the detectable 2CBA level was 61 mg kg^{-1}. Dahurian wild rye inoculated with a 1:1 mixture of *Pseudomonas aeruginosa* strain R75 and *P. savastanoi* strain CB35 was grown for 56 days in a growth chamber. Inoculating Dahurian wild rye with the mixed inoculum decreased 2CBA from an initial level of 61 mg kg^{-1} to 29 mg kg^{-1} 56 days after planting. Inoculating Dahurian wild rye had no effect on plant growth parameters or upon the bacterial community associated with the root surface. However, the inoculants increased rhizosphere soil's potential to degrade 2CBA, 3-chlorobenzoic acid but not 2,3-dichlorobenzoic acid or 2,5-dichlorobenzoic acid. In addition, inoculating Dahurian wild rye increased the enzymatic reduction of 2CBA levels in solution by over 300%. These results indicate that bacterial seed inoculants of Dahurian wild rye specifically increase the ability of the rhizosphere to degrade mono- but not di-chlorinated benzoic acids. This enhancement was not due to alterations in the rhizoplane community but may be related to an increase in enzyme expression in the rhizosphere.

PHA161 Algal Toxicity Testing of Antibacterial Agents Applied in Danish Fish Farming. Holten Lützhøft, H.-C.*; Halling-Sørensen, B.; S. E. Jørgensen, The Royal Danish School of Pharmacy, Copenhagen Ø, Denmark. Antibacterial agents are applied in intensive fish farming to prevent microbial infections among the fish. In Denmark, four tons of antibacterial agents are exposed directly to the aquatic environment along with the fish food pellets yearly - a quantity which is increasing. Only little knowledge exists about the environmental fate and effects due to the application of these chemicals in fish farming. The objective of this investigation was to establish algal toxicity data of the applied antibacterial agents. Algal growth inhibition tests were performed on seven antibacterial agents, amoxicillin, flumequine, oxolinic acid, oxytetracycline, sarafloxacin, sulfadiazine and trimethoprim using the test procedure described in the ISO-8692 protocol with *Microcystis aeruginosa* (freshwater cyanobacteria), *Rhodomonas baltica* (marine algae) and *Selenastrum capricornutum* (freshwater algae) as test organisms. EC_{50}-values obtained were in the range which could be found in the environment. EC_{50}-values for *M. aeruginosa* were in the range of 5-250 µg/L, whereas the EC_{50}-values for *R. baltica* and *S. capricornutum* were in the range of 2-300 mg/L. The results of this investigation showed that *M. aeruginosa* appeared to be two to three orders of magnitude more sensitive than both *R. baltica* and *S. Capricornutum* revealing a significant difference between procaryotic and eucaryotic algae.

PHA163 Reverse electron transport (RET) assay as a prescreening method for phytotoxicity. Schultz, E.*, Joutti, A., Vaajasaari, K. Finnish Environment Institute, Helsinki, Finland. Simple enzymatic *in vitro* toxicity tests are valuable tools for the rapid prescreening of environmental samples. RET assay is based on the measurement of NAD reduction catalyzed by the NADH-coenzyme Q reductase complex in submitochondrial particles. Since RET assay is sensitive to a wide range of chemical substances and it correlates well with animal toxicity tests, it was of interest to compare it with a plant growth test. Extracts from standardized leaching tests (TCLP, CEN, DIN, NEN) were tested for toxicity with RET and duckweed (*Lemna minor*) growth inhibition test. The samples industrial wastes (fly ash, metal working waste, jarosite) contained mainly inorganic substances, metals, heavy metals and some organic material (paper refinery waste, oil). Duckweed growth was measured as frond number, fresh weight and Chla. All samples except the paper refinery waste were toxic assessed by RET and duckweed. The RET assay was more sensitive than the duckweed test. The Chla measurement was more sensitive than the other plant parameters and it correlated with RET quite well. The sensitivity of RET was demonstrated in the five-step serial leaching test (NEN): the last two extracts were practically non-toxic when assessed by the duckweed test but RET was still significantly inhibited. Thus, RET used for prescreening is not likely to give false negative results. The extraction solution of TCLP was toxic as such for plants, while RET did not respond to it at the highest concentration tested. In conclusion: screening for toxicity with RET assay may be used for predicting aquatic animal and plant toxicity. These results may suggest a similarity for toxic mechanisms in animal and plant cells regarding essential biochemical reactions such as electron transport in mitochondria and chloroplasts.

PHA164 An Assessment of Boron Toxicity to a Phytoplankton Assemblage. Ziegenfuss, M.C.*, SmithKline Beecham Pharmaceuticals, King of Prussia, PA; Siesko, M.M., McLaren/Hart ChemRisk, Lake Charles, LA.; Goulden, C.E., Academy of Natural Science, Philadelphia, PA. This study was designed to investigate the impact of boron on the phytoplankton assemblage present in oxidation ponds of an industrial effluent treatment facility. The objectives were to determine the concentration of boron that: 1) causes a significant reduction in the growth of the resident phytoplankton; 2) may cause detrimental shifts in the dominant algae growing in the ponds; and 3) reduces photosynthetic oxygen production as a result of adverse growth effects. Determination of the impact of boron was made by a static 8-d bioassay in which an inoculum of oxidation pond algae was exposed in a geometric series of boron concentrations (0, 2, 4, 8, 16, and 32 mg B/L) prepared with filtered (0.4um) oxidation pond water. We used size-spectrum analysis by Coulter Counter® to determine changes in algal cell size, number, and total volume; visually identified oxidation pond phytoplankton algae to genus; and measured oxygen production of algae. At the end of the 8-d exposure, the no-observed-adverse-effect-concentration (NOAEC) for growth was 16mg B/L and the lowest-observed-adverse-effect-concentration (LOAEC) was 32mg B/L. No adverse changes in the algal assemblage composition were attributable to boron exposure. Shifts in species dominance from euglenoids to *Botryococcus* (a green alga) observed in test flasks in all test conditions were likely an artifact of the experimental method. Although algal growth was significantly altered at 32mg B/L, oxygen production was not adversely affected. Results of this study are presented in the context of a comparative risk assessment. The significance of evaluating an algal assemblage as opposed to a single species is discussed.

PHA165 Biomonitoring of overall herbicide toxicity to aquatic plants using two species of duckweed. Hatakeyama, S.* & Sugaya, Y., National Institute for Environmental Studies, Tsukuba, Ibaraki, Japan. For assessment of overall pesticide effects on river ecosystems, biomonitoring facilities were built in 1995 beside the Sakura River contaminated with various pesticides from the spring to summer. Growth (as total leaf-area) of two species of duckweed (*Lemna aoukikusa* and *Spirodela polyrhiza*) was measured on 4-, 7- and 14th-day after start of exposure to the river water using a flowthrough aquarium under continuous fluorescent light. The tests were conducted weekly or biweekly form April to August using the 10-young individuals of each species. Concurrently, *Selenastrum* growth tests (3-d) were conducted 3 times per week using water samples, and casual substances of its growth-inhibition was tested. Growth of the duckweed was almost completely suppressed from the mid-May to early June, and gradually recovered toward to the end of June, 1997. On the other hand, growth of *Selenastrum* recovered until early June, although its growth was severely suppressed in the mid-May by the combined toxicity of several herbicides. Photosynthetic activity on the leaf-surface of duckweed was also decreased in the period, when the growth of duckweed was intensively inhibited. Concentration of chlorophyll a in the river water also decreased in the period in which the growth of duckweed and/or Selenastrum was severely suppressed. It was considered that primary production might be significantly affected in rivers which are highly contaminated with several herbicides from the spring to summer.

PHA166 The Toxicity of Agricultural Pesticides to Phytoplankton and Subsequent Effects on Invertebrate Grazers. DeLorenzo, M.E.*; Pennington, P.L.; Fulton, M.H.; Scott, G.I., US Department of Commerce/NOAA, National Ocean Service, Charleston, SC. Although agricultural pesticide runoff in southeast coastal regions remains a critical issue, studies involving pesticide effects on phytoplankton are limited. Low-level chronic exposure to pesticides may impair primary productivity and thus alter important estuarine food sources at concentrations below which acute toxicity is expected. It is desirable to establish a link between direct effects at the microbial level and indirect effects on higher organisms. Such research will aid in creating a risk assessment model for estuaries based on the levels of pesticides detected. Two species of algae, *Selenastrum capricornutum* (green alga) and *Synechococcus* sp. (cyanobacterium), were exposed to two common agricultural pesticides, atrazine (a triazine herbicide) and endosulfan (an organochlorine insecticide). The experiments were 96-hour, static EC_{50} determinations, using direct cell counts and spectrophotometric absorbance at 664 nm as endpoints. Growth rates (divisions/day) were calculated based on cell densities and absorbance. Cell densities and absorbance values were strongly correlated. Cell densities at 96 hours were found to be a more sensitive measure of pesticide toxicity to phytoplankton than growth rate. Atrazine was more inhibitory to the algal species tested than endosulfan. The cyanobacterium species was more sensitive to pesticides than the green alga. Future research will focus on the joint effects of these compounds in a mixture, the potential interaction of these contaminants with nutrient enrichment, and the bioaccumulation of these compounds in phytoplankton and their subsequent effects in invertebrate grazers.

PHA167 The Effects of Pesticides and Nutrient Loadings in South Florida Canals on *Selenastrum capricornutum* and *Daphnia magna*. Pennington, P.L.*, DeLorenzo, M.E., Scott, G.I., Fulton, M.H., Key, P.B., Strozier, E.D., Chung, K.W., NOAA/NOS, Charleston, SC. Anthropogenic sources of nutrients and pesticides to surface waters has been an issue of concern in South Florida for many years. The purpose of this study was to evaluate the response of two standard bioassay organisms to water samples from South Florida canals. Water samples were collected from 5 sites within the C111 canal system in the South Florida Water Management District. Sampling sites were chosen based on associated land-use activities: agricultural, urban/suburban, and undeveloped. These samples were shipped to the National Ocean Service in Charleston, SC and were analyzed for pesticide residues (atrazine, endosulfan, and chlorpyrifos). These samples were also evaluated for toxicity using *S. capricornutum* and *D. magna*. The endpoints used for *S. capricornutum* bioassays were direct cell counts and absorbance at 664nm. Endpoints for *D. magna* included mortality and suppression of enzyme activity. Gas chromatographic methodologies revealed detectable levels of pesticides from sites within the C111 drainage canal system. Existing data (US EPA) indicate a history of high nutrient levels at several of the sampling sites. *S. capricornutum* showed varying growth responses to the water samples. Samples collected from sites with agricultural influences had significantly greater growth rates than controls. Conversely, exposure to a sample collected near an urban/developed area caused a significant depression in growth. No significant effects on survival or suppression of enzyme activity were observed in *D. magna* following exposure to any of the water samples. Ecotoxicological problems involving pesticides and nutrient loads are quite complex. Our future research will utilize laboratory tests to better delineate the individual and combined effects of nutrient inputs and pesticide loading in South Florida.

PHA168 Biochemical Responses of Estuarine Plants to Cd Exposure. Lytle, T.F.*, Lytle, J.S., Gulf Coast Research Laboratory, Ocean Springs, MS. Cadmium is a ubiquitous environmental toxicant of particular concern in estuarine areas impacted by sewage or industrial effluents. The estuarine plants, *Spartina alterniflora*, *Sesbania vesicaria*, and *Scirpus olneyi* have been cultured under greenhouse conditions and exposed to cadmium (methylmercury in an earlier experiment) through root uptake from soil spiked with solutions ranging up to10 μg/mL. Oxidative stress was denoted by increased loads of peroxidases, total ascorbic acid and glutathione within 2 days of initial exposure with eventual significant decrease in glutathione particularly in *S. vesicaria*. This drop in glutathione was further tested as possible activation of the enzyme responsible for conversion of glutathione to the metal sequestering group of compounds, phytochelatins. Phytochelatins are believed to function to detoxify metals, especially Cd, by sequestration through chelation. Plant tissues from exposed plants were digested in sulfosalicilic acid and extracts separated by HPLC using C18 reverse phase column with sequential elution in acetonitrile and identification of phytochelatins by post column. Levels of Cd-associated phytochelatins were notably evident in leaf tissues of all exposed plants with significantly more in those of *S. vesicaria*. In field validation of these measurements, *S. vesicaria* and sediments were collected in Biloxi Bay, Mississippi near sites where the NCAA Status and Trends noted significantly high and recently increasing levels of Cd in oyster, *Crassostrea virginica* with comparison to those in areas with undetectable Cd levels in sediments. Though statistically less reliable than laboratory experiments, plants in "high" Cd areas did evince oxidative stress and enhanced phytochelatins presumable due at least in part to presence of Cd in these sediments.

PHA169 Biomarkers of contamination in aquatic plants. Evidence of differential protein pattern in *Sagitaria sp.* exposed to Cd(II). Ramirez, S.; Porta, A. & Caffini N.O. Laboratorio de Ecotoxicología y Química Ambiental, Departamento de Ciencia y Tecnología, Universidad Nacional de Quilmes. *Sagitaria sp.* is an aquatic plant diffused along the Río de la Plata (Argentina) coast. This area shows important inputs of heavy metals as copper, cadmium and chromium due to massive industrial effluents discharges. The study of the biomarkers response in plants, as early indicators of pollution in aquatic systems, was recently proposed as a promissory method for application in biomonitoring programs. Among these indicators are emphasized phytochelatins and related peptides (PC), all cysteine-rich polypeptides (about 30% of its content), whose concentrations are increased in some plants exposed to heavy metals. In this work was studied the incorporation of Cd(II) by *Sagitaria sp.* and its effects on the electrophoretic protein patterns. *Sagitaria* specimens were cultured in Hoagland (1/10) media. with Cd(II) for 3 days. Controls were grown in the same conditions but without Cu(II). Temperature was fixed between 23 and 25 °C, and 16/8 h dark/ light cycles were adopted. The assays were conducted in duplicate. Cd (II) content was determined by atomic absorption spectrophotometry, and the protein pattern modification was followed by denaturing electrophoresis (SDS-PAGE). In SDS-PAGE gels bands at 10 kDa shown a significant increase. These results let us to suppose that *Sagitaria sp.* could be a sensitive and usefully sentinel in aquatic biomonitoring programs, using the differential protein pattern as indicator of heavy metal exposure.

PHA170 Effects of Copper Sulfate on *Typha latifolia* Seed Germination and Early Seedling Growth in Aqueous and Sediment Exposures. Muller, S.L.*, Clemson University, Pendleton, SC., Huggett, D.B., University of Mississippi, Oxford, MS; Rodgers, J.H., Jr., Clemson University, Pendleton, SC. The vascular macrophyte *Typha latifolia* is a potential candidate for determining phytotoxicity in aquatic systems. *Typha latifolia* seed germination, shoot growth, and root elongation were evaluated in 7d aqueous exposures with copper sulfate (nominal concentrations of 12.5, 25, 50,100, 200, and 400 µg Cu/L). Mean measured aqueous copper concentrations were 10, 23, 41, 62, 174, 402 µg Cu/L and were ≥ 62% of nominal concentrations. Observed seed germination and seedling shoot growth were not significantly affected as compared to controls. Mean no-observed effect concentration (NOEC) and lowest observed effect concentration (LOEC) for root elongation were 20.8 µg Cu/L and 41.7 µg Cu/L, respectively. 7d sediment experiments were also conducted by amending University of Mississippi Field Station sediments with copper sulfate to nominal concentrations of 6.25, 12.5, 25, 50,100, and 200 mg Cu/kg. Mean measured sediment copper concentrations were 7.9, 17.1, 21, 51.2, 89.5, and 173.5 mg Cu/kg and were ≥ 84% of nominal concentrations. Observed seed germination was not significantly different from controls. However, mean NOEC and LOEC for seedling shoot growth were 100 mg Cu/kg and 200 mg Cu/kg, respectively, and mean NOEC and LOEC for root growth were 10.4 mg Cu/kg and 20.8 mg Cu/kg, respectively. These results demonstrate that *Typha latifolia* can be successfully utilized for assessing aqueous and sediment phytotoxicity.

PHA171 The effect of size and root age on the toxicity of simazine to *Myriophyllum aquaticum* and *Canna hybrida*. Knuteson SL*, Wilson PC and SJ Klaine, Clemson University, Pendleton, SC.Pesticides are used to improve the quality of our lives and surroundings. However, improper pesticide use can also lead to environmental problems such as water quality degradation and ecological stress. Recent research in our laboratory has focused on the development of constructed wetlands to assimilate pesticide contaminated water. For improved aesthetics, these wetlands have been established with ornamental plant species. The effectiveness of a plant species for phytoremediation depends in part on its tolerance to the pesticide. Plant tolerance to pesticides may vary depending on plant size and age. This study examined the influence of plant size and root age on the toxicity of the herbicide simazine to an ornamental wetland plant, *Myriophyllum aquaticum* (Parrot feather) and *Canna hybrida* (King Humbert). Plants were exposed to concentrations of 0, 0.06, 0.18, 0.36, 0.61, and 1.49 mg/L simazine in 10% HoaglandÕs nutrient media. Toxicity endpoints were 7-d biomass production during exposure and post exposure periods (increases in fresh weight and length), offshoot production and water use. Smaller, newly rooted *M. aquaticum* were more sensitive than the larger plants with well established roots. Results suggest that performance of wetlands constructed for simazine remediation would increase as they age.

PHA172 Toxicity and Detoxification of Trichloroacetic Acid in the Aquatic Macrophytes *Myriophyllum spicatum* and *Myriophyllum sibiricum*. Hanson, M.L.*, Jacobs, D., Solomon, K.R., Sibley, P.K., Centre for Toxicology, University of Guelph, Canada; Muir, D.C.G., National Water Research Institute, Burlington, Canada; Mabury, S.A., Dept. of Chemistry, University of Toronto, Canada. Trichloroacetic acid (TCA) is becoming increasingly ubiquitous in the aquatic environment. Sources of TCA into natural water systems include herbicide use, as a by-product of water disinfection, from volatilization into air from spent bleach liquor of kraft pulp mills, and as a natural fungal product. This compound is phytotoxic and has been used to control aquatic macrophytes, including *M. spicatum*. In laboratory evaluations, chlorophyll a/b and carotenoid concentrations appeared to be the most sensitive endpoints with reductions of 40, 50, and 33 percent from the controls respectively in *M. spicatum* at 1000μg/L. In *M. sibiricum*, there was a 12, 3, and 21 percent reduction in these endpoints respectively at 1000μg/L. *M. spicatum* had a 26 percent reduction, while *M. sibiricum* had a 20 percent increase in total length at 1000μg/L. Laboratory data indicates that *M. spicatum* is the more sensitive of the two species to TCA. During the summer of 1998, *M. spicatum* and *M. sibiricum* will be exposed to trichloroacetic acid at the Guelph Microcosm Facility and allowed to develop over the field season. The microcosms hold 12m^3 of water and are designed to replicate natural aquatic ecosystems. Plants will be removed at regular intervals and assessed for growth, chlorophyll a/b and carotenoid concentrations, wet and dry weight, root length, membrane permeability, and glutathione levels. At the end of the field season plants will be removed and assessed for total biomass. The results of these experiments will be presented and discussed.

PHA173 Effects of Picloram in Irrigation Water on Soybean Growth and Final Yield. Schwab, D., Lee, K. B. ABC Laboratories, Columbia, MO; Wright, J. P. *, Hanselman K. A., Robb, C. K., Dow AgroSciences LLC, Indianapolis, IN. Field studies were performed during the past two growing seasons to determine the effects of picloram in irrigation water. Using an overhead sprinkler irrigation method, treatments were applied to soybeans at concentrations of 0.5 and 2 parts per billion (ppb) in 1996. Failing to measure any effects at these low rates, higher concentrations of 5, and 20 ppb were made in 1997. Application rates were verified by chemical analysis. In both years, applications were made at three different crop growth stages, V2 (between emergence and appearance of the 2nd node), R2 (full bloom) and R4 to R5 (beginning seed). The control plots were irrigated concurrent with the applications to the treated plots. The volume of water applied at each application ranged from 0.5 to 1 inch of water per acre. Visual injury ratings were taken at weekly intervals beginning approximately 1 week after the first application and continuing to approximately the R7 growth stage (beginning maturity). Soybean grain harvest occurred at crop maturity. No visible injury was observed at 0.5, 2, and 5 ppb, but slight leaf curling was observed at 20 ppb. There was no significant difference in final grain yields between the treated and control plots.

PHA174 Impact of Anthracene Derivatives on the Aquatic Higher Plant Lemna gibba: Photosynthetic Activity and Molecular Shape Modelling. Ali Mallakin, Brendan J. McConkey, Sudhakar T. Babu,, Paul Mezey*, D. George Dixon, Bruce M. Greenberg, Dept. of Biol., Univ. of Waterloo, * Dept of Chem., Univ. of Saskatchewan, Saskatoon, Canada. Photochemical reactions in sunlight are a common route of modification ofaromatic contaminants in the environment. For example, the toxicity of polycyclic aromatic hydrocarbons (PAHs) is enhanced by actinic radiation via photosensitization (e.g. production of active oxygen) and by photomodification of the chemicals to more toxic species. Photomodification of aromatic contaminants results in more electrophilic and hence more reactive compounds. In this study we have determined the relative toxicity of different anthracene derivatives. HPLC analysis of anthracene oxidation products revealed many of the products are quinones, phenols and benzoic acids. Toxicity was assayed as inhibition of growth and impairment of photosynthesis in Lemna gibba (duckweed) performed under photosynthetically active radiation (PAR) and simulated solar radiation (SSR). Chlorophyll a fluorescence quenching was found to be the most sensitive indicator of effects of polycyclic aromatic hydrocarbons on plants. Using photosynthetic activity due to electron transport downstream from PSII we were able to rank the relative toxicity of 12 ANT photoproducts. Experiments with PAR and SSR showed that the phytotoxicity of these chemicals is photoactivated, with UV radiation being the most influented spectral region in enhancing toxicity. Finally, a new approach for modeling toxicity of these chemicals has been applied by using the molecular shape of anthracene derivatives. Electron density analysis for the 12 ANT photoproducts has been completed. The empirical toxicity model has been trained to explain the toxicity of the tested compounds and to predict the toxicity of a wide group of organic hydrocarbon contaminants.

PHA175 Influence of adsorbed fulvic acid on algal metal uptake. Vigneault, B.* and Campbell, P.G.C., INRS-Eau, Université du Québec, Sainte-Foy, QC, Canada. According to the Free-Ion Model (FIM), algal uptake of a dissolved metal should be proportional to the free-metal concentration in solution. However, the FIM has not yet been validated with the dominant ligands found in natural waters (fulvic/humic acids), nor does it account for the possible effects of these molecules when directly adsorbed at the biological surface (see *Can.J.Fish.Aquat.Sci.* 54: 2543-2554 (1997)). To test the applicability of the FIM in the presence of such natural ligands, we are using a unicellular green alga (*Selenastrum capricornutum*) as the test organism, Cd as the test metal, nitrilotriacetate (NTA) as the synthetic ligand, and well-characterized Suwannee River fulvic acid (FA) as a model natural ligand. Cadmium uptake experiments were designed to compare uptake-exposure relationships observed in the presence and absence of FA. In both cases algal cells were exposed to a series of increasing Cd^{2+} concentrations; in one case $[Cd^{2+}]$ was controlled with a Cd-NTA buffer, in the other case in a Cd-FA acid system. Free Cd^{2+} concentrations were both measured by an equilibrium ion-exchange technique and calculated. After exposure the cells were washed with EDTA to remove surface-bound metal, and the intracellular Cd content was determined. At pH 7, as predicted by the FIM, the Cd uptake is similar in the NTA and FA systems. However, at pH 5, Cd uptake (over the same range of free Cd^{2+} concentrations) is markedly decreased in presence of fulvic acid compared to the NTA system. Since adsorption of fulvic acid at the *Selenastrum* cell surface is measurable at pH 5, but not at pH 7, we suspect that adsorbed FA affords additional protection to the alga at pH 5.

PHA176 Quantification of Organic and Inorganic Selenium Compounds in the aquatic plant *Ruppia maritima*. F.A. Hix* and F.C. Bailey, Middle Tennessee State University, Murfreesboro, TN. The aquatic plant *Ruppia maritima* (wigeongrass) is known to readily accumulate selenium (Se) and as a known waterfowl food, represents a means of food chain transfer of Se. The objectives of this study were to investigate the potential of *R. maritima* to accumulate Se in both organic and inorganic forms and classify the plant as a selenium accumulator or nonaccumulator species. Organic forms of Se, such as selenoamino acids, are generally more toxic to wildlife than inorganic forms. Selenium accumulator plants apparently avoid Se toxicity by "shunting" Se into selenium-containing amino acids which are not incorporated into proteins, such as selenocystathionine. Plants dosed with sodium selenite, sodium selenate, or seleno-dl-methionine at 1 mg/L as Se accumulated Se to 73, 114 and 321 µg Se /g dry weight respectively. Ethanol extracts of plants were subjected to cation exchange chromatography to separate selenoamino acids from inorganic Se. The organic and inorganic extracts, residue left after ethanol extraction, and cation exchange columns were analyzed for Se by hydride generation atomic absorption spectrometry (HG-AA). Organic and inorganic Se levels varied depending on the Se species dosed. The ratios of organic: inorganic: residue: column Se fraction by treatment were: sodium selenite- 43.0, 11.2, 23.8, 22.0%; sodium selenate- 27.0, 16.1, 39.2, 17.2%; seleno-dl-methionine- 13.4, 27.9, 49.9, 9.7%. Analysis of ethanol extracts by paper chromatography and HG-AA indicate the presence of selenomethionine in all the Se treatments and selenocystathionine in the selenate treatment extract. Because of the high level of Se accumulated in these plants and the fact that selenocystathionine is typically found only in small amounts in nonaccumulator plants, *R. maritima* may therefore be classified as an aquatic secondary selenium accumulator.

PHA177 Green algae growth recovery studies after pesticide exposition. Sáenz, M.E., W.D. Di Marzio, J.L. Alberdi* and M.D.C. Tortorelli, Ecotoxicology Research Program, Dept. of Basic Sciences, National University of Luján (Bs. As., Argentina). Growth recovery studies were conducted in order to assess the level of dammage caused by pesticides to natural phytoplankton populations.Pesticides may cause algistatic effects which allows a recovery of algae populations, when harmful concentrations are not more presents. Pesticides may kill algae cells, not allowing population recovery even when letal concentrations have desappeared from water column. Pesticides commercial formulations used were the herbicides Osaquat (Paraquat) and Round-up (Glyphosate) and the pyrethroid Baytroid (Cyfluthrin). These products are used in agricultural practise in the Buenos Aires Province, entering to aquatic systems by surface runoff or spray drift. Toxicity test of 96 hrs of exposition were conducting using as test organisms Selenastrum capricornutum, Scenedesmus quadricauda, Scenedesmus acutus and Chlorella vulgaris. The general design of USEPA (1989) was used for the estimation of toxic effects. 96 hs EC 50 were calculated. The procedure of Payne and Hall (1979) was used to determine algistatic or algicidal effects, after a recovery period of ten days. Osaquat algistatic concentrations for algae species were 1.02, 0.63, 0.12 and 0.12 mg Pq/L, respectively. Round-up caused algicidal effects at concentrations between 10 and 100 mg Gly/L. Baytroid caused algicidal effects at 4.6 mg Ci/L to S. quadricauda and S. acutus and at 12.3 mg Ci\L to Selenastrum capricornutum and Chlorella vulgaris. Populations of different algae species affected by pesticides were analysed. The level of potencial dammage to algae natural populations were discussed.

PHA178 The Effects of PCB Congeners on Dopamine Levels and the Dopamine Metabolites DOPALD and DOPAC in PC12 Cells. Pivorun, E.B.*, Williams, R. G., Montie, E.W., Clemson University, Clemson, SC. Although studies have demonstrated that PCB congeners reduce dopamine (DA) levels within the CNS and DA within PC12 cells, the mechanism(s) leading to these reductions is unknown. Although, it has been hypothesized that PCB exposure inhibits DA synthesis, there is no convincing evidence. Our studies with ortho-substituted PCB congeners suggest that alterations in DA levels are dependent on both an increase in the extracellular secretion of DA and metabolism of DA via monoamine oxidase (MAO). HPLC with electrochemical detection was used to monitor the levels of intra- and extracellular DA and its metabolites: 3,4 dihydroxyphenylacetaldehyde (DOPALD), 3,4-dihydroxyphenylacetic acid (DOPAC), and 4-hydroxy-3-methoxyphenylacetic acid (HVA). All observations are reported relative to pargyline (an inhibitor of MAO activity) treated cells; this provides baseline levels of DA and DA metabolites prior to experimental incubations of 30 and 60 min duration. Control cells were incubated with DMSO and PCB-treated cells were incubated with 10ppm PCB congener (2,2'; 2,2',4,6; or 2,2'4,4') in DMSO. Incubation in the presence of these congeners resulted in a selective increase in the extracellular levels of DOPALD, the initial metabolite of DA produced via MAO activity. There were major differences in the effects of the congeners. The 2,2',4,6 congener elicited a more profound elevation in DOPALD relative to the 2,2' and 2,2'4,4' congeners. On average the control cells displayed an elevation in DOPALD of 450% relative to the pargyline treated cells (60 min). At 60 min the 2,2',4,6, the 2,2', and the 2,2'4,4' congeners elicited an 800%, a 600% and a 450% increase in DOPALD, respectively. Although intracellular DA levels were depressed and the extracellular DA levels elevated by the three congeners, these elevations were relatively minor compared to the elevations in DOPALD levels. HVA levels which are controlled by catechol-O-methyltransferase activity were not markedly affected by PCB exposure.

PHA179 Hepatic bioindicator enzymes in a sentinel species (*Chrysemys picta*) from Cape Cod, Massachusetts: Seasonal, sex and location related differences. Rie, M.T.*, Lendas, K.A., Boston University, Boston, MA; Woodin, B., Stegeman, J.J., Woods Hole Oceanographic Institute, Woods Hole, MA; Callard, I.P., Boston University, Boston, MA Here we report changes in biomarker enzymes and protein expression in *Chrysemys picta*, as part of a study of potential xenobiotic impacts. In animals from Moody Pond, a site potentially impacted by xenobiotics, rates of hepatic microsomal EROD activity (a CYP1A activity) were very low in both sexes in early Spring; female values peaked in early July (6.8±1.4 pmol/min/mg) and male values peaked in June (8.82±2.1 pmol/min/mg). EROD rates in females were significantly higher than in males in early July (ANOVA; $p<0.05$). At the candidate clean site, Washburn Pond, EROD rates in males peaked in May (4.2 pmol/min/mg), and in females in early July (2.5 pmol/min/mg). There were sex differences seen in May (t-test; $p<0.05$). There were differences in EROD rates between turtles from the two locations (ANOVA; $p<0.05$). Western blot analysis with mouse anti-scup monoclonal antibody showed that levels of CYP1A protein were also detected and followed a similar pattern of expression as EROD activity. Glutathione-S-Transferase (GST) activity also showed a seasonal pattern at both sites. In Moody Pond, values for both males and females were highest in June (26.6±4 x 10-4 umol/min/mg (females) and 20.23±4 x10-4 umol/min/mg (males)), with male values steadily declining through October and female values showing no significant change. In Washburn Pond, values for both males and females are high in May, drop to a basal level in June/July and rise again in September. There was no significant difference in activity between males and females, but there were significant differences between the two sites, with activity being higher at the potentially impacted site. Supported by ES07381 to IPC and JJS.

PHA180 Dietary Accumulation and Biochemical Responses of Juvenile Rainbow Trout *(Oncorhynchus mykiss)* Exposed to PCB-126. Fisk, A.T.*, University of Manitoba, Winnipeg, MB, Canada; Brown, S.B., Muir, D.C.G., National Water Research Institute, Burlington, ON, Canada; Metner, D.A., Lockhart, W.L., Freshwater Institute, Winnipeg, MB, Canada. Juvenile rainbow trout were exposed to 2 dietary concentrations (12.4 and 126 ng·g^{-1}) of polychlorinated biphenyl congener 126 (CB 126) for 30 days followed by 160 days of clean food to assess bioaccumulation and biochemical responses (thyroid hormones, liver vitamins (retinoids and tocopherol) and liver ethoxyresorufin-O-deethylase EROD)) to this non-ortho substituted PCB. The half lives of CB 126 in the rainbow trout ranged from 82 to 120 d and biomagnification factors (BMF) ranged from 2.5 to 4.1. EROD activity was elevated in rainbow trout exposed to the high concentrations by day 5 of the uptake period when liver concentrations were 4.3 ± 1.4 ng·g^{1}, and remained elevated through day 160 of depuration despite liver concentrations falling to 0.95±0.78 ng·g-1. EROD activity in the low exposure rainbow trout was elevated by day 30 of uptake and remain above control levels 40 day after cessation of exposure. The half life and BMF of CB 126 are greater than other ortho-substituted PCBs using similar experimental protocols, confirming the known potential for this CB congener to biomagnify in aquatic food chains. The shorter CB 126 half life observed in rainbow trout exposed to higher concentrations may be related to greater EROD activity and subsequent biotransformation of CB 126 in these fish.

PHA181 Responses of Hepatic Induction and Lesions Caused by β-Naphtoflavone, Pyrene, Pyridine, Phenobarbital and Sediment Extract in Adult Tilapia *(Oreochromis niloticus)*. Zapata-Pérez, O *., Simá-Alvarez, R., Noreña-Barroso, E., Gold-Bouchot,G., Albores-Medina, A. CINVESTAV-IPN. Mérida, Yucatán, México. Tropical fish Tilapia (*Oreochromis niloticus*) received a single i.p. injection dose of β-naphtoflavone, pyrene, pyridine, phenobarbital (equivalent to20 mg kg^{-1} of body weigth) and sediment extract from Chetumal Bay. Fish were sacrifed 24 hr after injection and hepatic levels of Cytochrome P-450, and EROD, MROD, BROD, PROD and ρ-NPHL activities were measured as well as damages caused in organs such as gills, liver and spleen, using standard histological tests. Fish treated with pyrene, the sediment extract and β-naphtoflavone increased the P450 content with respect to fish control, with values of 0.057, 0.121 and 0.166 nmol mg protein^{-1}, respectively. EROD activity was inhibited in 24 h in the group treated with pyrene, but increased (4-fold) in fish inyected with sediment extract and (10-fold) in the β-naphtoflavone treated group. MROD activity did not increase as much with pyrene as it did with sediment extract (4-fold) with respect to the control. On the other hand, the sediment extract increased the MROD activity in the same magnitude as the EROD activity. BROD activity was inhibited with the sediment extract and no activity was detected for the rest of treated groups. In this study we did not detect PROD and ρ-NPHL activities.

PHA182 Hepatic Microsomal Monooxygenase Activity and Histological Lesions Caused by Pyrene in Tilapia (*Oreochromis niloticus*). Zapata-Pérez, O *., Noreña-Barroso, E., Simá-Alvarez, R., Güemez, J., Gold-Bouchot,G., Albores-Medina, A. CINVESTAV-IPN. Mérida, Yucatán, México. In the present study the hepatic activity of a microsomal P-450-dependent monooxygenase was determinated in tilapia (*Oreochromis niloticus*). The effect of pyrene in the relative cytochrome P-450 content and EROD activity was evaluated, as well as the damages caused in blood cells and gills, liver and spleen as a result of the presence of this compound in the organisms, using standard histological procedures. Male adult fish were injected with a single i.p.dose of pyrene (20 mg kg^{-1}), and fish were sacrificed at 1, 3, 5 and 7 days after the injection. The control value of P-450 content was 0.049 nmol mg of protein^{-1}; P-450 increased from 46 % in day 1 up to 417 % in day 7. EROD activities showed a different pattern than P-450. EROD activity decreased in day 1 (10.9%), at day 3, 5 and 7 increased 8.4%, 123.6% and 473.6%, respectively. The blood and histological observations did not show abnormalities in the control fish and we detected that 90% of the organisms injected with pyrene presented lesions. Damages in gills such as hyperplasia, hypertrophy and edema were found. The liver observations showed the presence of basophilic cells, necrosis, hyperemia, cell degeneration and inflamation.

PHA183 Interaction of Tribuyltin with 3,3',4,4',5-pentachlorobiphenyl - Induced Ethoxyresorufin-o-deethylase (EROD) Activity in Rat Hepatoma Cells. Kannan, K.*, Villeneuve, D., Blankenship, A., Giesy, J.P., Michigan State University, East Lansing, MI. Tributyltin (TBT) is an effective biocide that enters the aquatic environment mainly from its employment in antifouling paints on vessels. TBT has been shown to interact with hepatic cytochrome P4501A leading to the destruction of the native enzyme and its activity in fish. In this study, interaction of TBT with a classical P4501A inducer, 3,3',4,4',5-pentachlorobiphenyl (PCB-126), on EROD activity was examined *in vitro* using H4IIE - bioassay. TBT and PCB-126 were exposed, individually or in combination, at environmentally realistic concentrations to H4IIE cells. TBT was cytotoxic at concentrations greater than 98 nM. Co-exposure of PCB-126 enhanced the cytotoxicity of TBT in the 24-98 nM range. EROD activity in H4IIE cells was significantly increased by exposure to PCB-126, and this effect was potentiated by coexposure to small, non-cytotoxic concentrations of TBT. The induction of CYPIA activity in the presence of both an inducer (PCB-126) and small concentrations of an inhibitor (TBT) indicates that TBT does not interfere with the Ah receptor binding, but acts at the transcriptional level. Potentiation of EROD activity and cytotoxicity as a consequence of co-exposure to PCB- 126 and TBT is of considerable toxicological significance, given their co-accumulation in a variety of aquatic organisms.

PHA184 Tributyltin (TBT) Potentiates 3,3',4,4',5-Pentachlorobiphenyl (PCB-126)-Induced Hepatic CYP1A Activity in Channel Catfish, *Ictalurus punctatus*. Rice, C.D.*, Roszell, L.E. CEHS, College of Veterinary Medicine, Mississippi State University, P.O. Box 9825, Mississippi State, MS 39762. Planar halogenated aromatic hydrocarbons (HAHs) and polycyclic aromatic hydrocarbons (PAHs) are found in many aquatic systems. Their toxic effects are initiated primarily through the cytosolic aryl hydrocabon receptor (Ahr). However, only rarely are Ahr-binding contaminants found alone in the environment. Tributyltin (TBT), a common antifouling biocide, is also found in many harbor estuaries and their tributaries. Several reports indicate that TBT inhibits the cytochrome P4501A system of fish, at least in vitro, but our previous studies with rodents indicate that TBT potentiates PCB-126 (PeCB)-induced hepatic CYP1A in vivo. We exposed channel catfish, Ictalurus punctatus, to PeCB, TBT, or both in combination, with corn oil (CO) serving as the carrier control. Immunoreactive hepatic CYP1A protein and EROD activity were measured 3 and 7 days after (1) a single dose of 0.01, 0.1, or 1 mg/kg of each or both in combination, and (2) as six injections of 0.017, 1.7, or 17 ug/kg of each (or in combination) given at 72 hr intervals over a 16 day period to yield a cumulative dose of 0.01, 0.1, or 1 mg/kg. When administered alone, PeCB, but not TBT, greatly induced these two CYP1A parameters. When given in combination TBT (0.01 and 0.1 mg/kg), potentiated PeCB-induced activity. This property of TBT was even more pronounced in the fractionated-repeated exposure study. Moreover, EROD activity did not always reflect CYP1A protein induction in the repeated exposure study; enzyme activity was inhibited by TBT at doses that potentiated PeCB-induced

CYP1A protein (0.01 and 0.1 mg/kg of TBT). In summary, TBT potentiates PeCB-induced CYP1A in channel catfish at doses considered to be environmentally relevant. (Funded by EPA-R822364010).

PHA185 MFO Induction of Three Australian Fish Species. Smith B.J.*, RMIT University, Melbourne, Australia, and Gagnon M.M., RMIT University, Melbourne, and Curtin University, Perth, Australia. The use of mixed function oxidases (MFOs) as a biomarker of pollution in Port Phillip Bay was investigated in three indigenous fish species, sand flathead *(Plathycephalus bassensis)*, blue throat wrasse *(Notalabrus tetricus)* and sixspine leatherjacket *(Meuschenia freycineti)*. Investigation of the relative MFO induction potential was performed by intraperitoneally injecting each experimental fish species with 100 µg/kg of 3,3',4,4',5-pentachlorobiphenyl (PCB 126) using corn oil as a carrier. Hepatic induction was fluorimetrically measured by the activity of 7-ethoxyresorufin 0-deethylase (EROD) 10 days post-injection. Of the three species tested, the sand flathead was selected as the most suitable indicator species for its high MFO induction potential and desirable characteristics related to its population biology. Bluethroat wrasse appeared relatively uninducible by PCB 126, while sixspine leatherjacket demonstrated a medium induction potential but was unsuitable as a biomonitoring tool because of its scarcity and distribution limited to reef beds.

PHA186 PAH bile metabolite, hepatic EROD activity and BUN levels in Ottawa River white suckers and common carp: comparisons between two sampling years. Cormier, S.M., Lin, E.L.C., U.S. EPA, Cincinnati, OH; Millward, M.R., PAI-SAIC/SBI, Cincinnati, OH; Schubauer-Berigan, M., Subramanian, B.*, U.S. EPA, Cincinnati, OH; Counts, B., Altfaler, D., Ohio EPA. Benzo[a]pyrene (BAP)- and naphthalene (NAPH)-type, polycyclic aromatic hydrocarbon (PAH) bile metabolite levels, hepatic ethoxyresorufin-*O*-deethylase (EROD) activity and blood urea nitrogen (BUN) level were measured in white suckers (*Catastomus commersoni*) and common carp (*Cyprinus carpio*) taken during 1993 and 1996 Ohio EPA sampling in the Ottawa River. The river is heavily impacted by multiple point source inputs in and around Lima, Ohio. Comparisons showed that levels of both bile metabolites and EROD activity were generally elevated downstream relative to an upstream reference site, especially around the clustered inputs in and immediately downstream of Lima, OH. When levels of the PAH bile metabolites from fish from each site were compared to estimated reference/criteria values, calculated following the recommendations of the International Federation of Clinical Chemistry, bile metabolite levels were found to be in excess of the criteria values at most downstream sites. BUN levels did not show clear spatial patterns. Comparisons were also made between years for sites sampled in 1993 and in 1996 to determine if changes could be detected over time. Results varied between the species. Comparisons between years indicated that, though levels of all biomarkers were still elevated relative to the upstream site and calculated criteria values, they were significantly lower at many sites in 1996 than in 1993 and higher at no site, suggesting possible improvement in river conditions over the period.

PHA187 Induction of Hepatic Ethoxyresorufin-O-Deethylase (EROD) in Rainbow Trout (*Oncorhynchus mykiss*) Exposed to 3,3',4,4'-Tetrachlorodiphenyl Ether by Intraperitoneal Injection or Gavage Intubation. G. M. Pastershank[1]*, C.D. Metcalfe[2], and Y. Kiparissis[2]. [1] University of Ottawa, Ottawa, ON, K1N 6N5. [2] Trent University, Peterborough, Ontario, K9J 7B8 Canada. The induction of hepatic ethoxyresorufin-O-deethylase (EROD) was studied in rainbow trout (*Oncorhynchus mykiss*) exposed to 3,3',4,4'-tetrachlorodiphenyl ether (3,3',4,4'-TCDE) by two routes of administration. A single dose of 3.3 μmol Kg^{-1} of 3,3',4,4'-TCDE was administered either by intraperitoneal (i.p.) injection or gavage intubation and fish were sampled over a duration of 24 d post-exposure. Gavage intubation of the test compound caused significantly higher induction of EROD enzyme activity than the controls at 3, 12 and 24 d post-exposure. However, the degree of EROD induction was weak at 2.9 ± 0.5, 2.9 ± 0.4 and 3.4 ± 0.3-fold higher than activity in control fish, respectively. These levels of EROD induction are at least 10-fold lower than those in similar studies with comparable doses of 2,3,7,8-TCDD and 3,3',4,4'-tetraCB. No significant EROD induction was observed in fish administered 3,3',4,4'-TCDE by i.p. injection. The results of this study show that the routes of exposure can influence the degree of hepatic EROD induction.

PHA188 Comparison of Dioxin Toxic Equivalents in New York Harbor Sediments by P450 Reporter Gene System and EPA Method 8290. Anderson, J.W.*, and Jones, J.M. Columbia Analytical Services, Carlsbad, CA., Reiss, M.R., U.S. Army Corps of Engineers, New York District, New York, NY., Peven, C. S., Battelle Ocean Sciences, Duxbury, MA, and Schrock, M.E. Battelle Laboratories, Columbus, OH. Twenty-three sediments which had previously been characterized for the content of dioxins and furans by method 8290 were shipped blind to Columbia Analytical Services for extraction (EPA method 3550) and testing with the biomarker assay, P450 RGS. Extracts (2 L of 1:20 dilutions) were tested at both 6 and 16 hours of exposure to this transgenic cell line to determine the relative contributions of PAHs and dioxins in the samples. 6-hr responses were most often 4 to 6 times greater than the 16-hr responses, indicating that significant amounts of PAHs were present. RGS responses at 16 hr were used to calculate RGS Toxic Equivalents from a standard curve, where pg/mL of a standard mixture of 17 dioxins and furans demonstrated a 1 to 1 relationship to fold induction of the cells. After omitting one suspect sample of the 23, the TEQs obtained using the two methods were well correlated (r^2=0.81). The slope of the regression indicated that RGS TEQs were approximately 200 times higher than those from chemical analyses and suggested that the significant levels of PAHs present in the sediments contributed to the response. The high correlation of TEQ values by the two methods shows that the P450 RGS method can be used, with chemical confirmation on the order of 20% of total samples, to estimate TEQs and identify priority samples while realizing significant cost and time savings in sediment contamination assessment scenarios.

PHA189 Relative Potencies of Halowax Mixtures and Individual Polychlorinated Naphthalenes in H4IIE-Luciferase Cells. Blankenship, A.L.*, Kannan, K., Villalobos, S.A., Giesy, J.P, Michigan State University, East Lansing, MI; Falandysz, J., University of Gdansk, Gdansk, Poland. Polychlorinated naphthalenes (PCNs) are nearly ubiquitous environmental pollutants that are structurally related to polychlorinated dibenzo-p-dioxins (PCDDs), dibenzofurans (PCDFs), and biphenyls (PCBs). Despite being nearly ubiquitous, little is known about the fate, transport, and biological effects of individual PCN congeners. The purpose of the current study was to utilize an H4IIE-luciferase bioassay to determine relative potencies for 20 individual PCNs (there are 75 possible congeners) and 6 Halowax mixtures compared to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) as a standard reference compound. Individual PCN congeners were tested for both their ability to produce a statistically significant response relative to the solvent control and their ability to elicit a full dose-response curve. The most potent congeners were 1,2,3,4,6,7-HexaCN, 1,2,3,5,6,7-HexaCN, and 1,2,3,4,5,6,7-heptaCN, with relative potencies of 0.0039, 0.001, and 0.001, respecitively, compared to TCDD. As expected, significant structure-activity relationships were observed. In general, full dose-response curves were obtained for more chlorinated congeners (e.g., penta-, hexa-, and hepta-CNs) whereas many of the lesser chlorinated congeners as well as octa-CN were generally inactive. The position of chlorines in hexa- and hepta-CNs is also an important determinant of activity. In general, lateral substitution is an important determinant of activity, but not sufficient, as illustrated by the inactivity of 2,3,6,7-tetraCN to cause Ah receptor-mediated activity. Of the Halowax mixtures tested, the relative potencies were: 1051 > 1014 > 1013. Halowaxes 1000, 1001, and 1099 were inactive in the H4IIE-luciferase bioassay. The relative potencies were applied to literature-derived data on concentrations of PCN congeners in Halowaxes and environmental mixtures to assess the potential contribution of PCNs to total TCDD equivalents (TEQs) in complex mixtures.

PHA190 CYP1A Expression in Fish from a Estuarine Zone in South of Brazil. Bainy, A.C.D.*, Mezzari, M.P., UFSC, Florianopolis, SC, Brazil; Hostim-Silva, M., Univali, Itajai, SC, Brazil; Marques, M.R.F., UFSC, Florianopolis, SC, Brazil. Monitoring programs of aquatic environments established in North America and Europe have included the analysis of biomarkers in order to get more data about the effects of the pollutants upon the organisms. In Brazil, as well as in other Latin American countries, several contamination cases have been observed around the coastal zones, particularly in the estuarine areas, but scarce information is available about the biological effects of pollutants discharged in the environment. The estuarine complex of Itajai-Açu River, located in Santa Catarina State, south of Brazil, is a very important economic region which possesses several industries, hosts an harbor and hold a very important fishing industry. Because of that, this estuary has periodically received the impact of contaminants that may be affecting the health of the aquatic organisms, as well as the ecosystem. The main purpose of this study was to verify the expression of the routinely used biomarker of organic pollution, cytochrome P450 isoform (CYP1A), in liver of fish caught in different sites of this estuary. Three species of sediment associated fish, *Stellifer rastrifer* (Scienidae), *Citharichthys spilopterus* (Bothidae) and *Catathyridium garmani* (Achiridae) were collected monthly by a bottom trawling net, for 20 min. Liver samples were immediately extracted and immersed in liquid nitrogen. The samples were homogenized and centrifuged in order to obtain the microsomal fraction. The proteins were separated in SDS-PAGE 12 % and transferred to a nitrocellulose membrane. Immunoblot analysis was performed using as primary antibody, the monoclononal Ab 1-12-3 against scup P450E (CYP1A), gently donated by Dr. John J. Stegeman, and as secondary antibody, the alkaline phosphatase-(AP) conjugated goat anti mouse IgG (Biorad). We detected significant CYP1A levels in all analyzed species. Based on these results, we conclude that the fishes from the estuarine complex of Itajai-Açu River are being exposed to Ah-receptor agonists. Further ecotoxicological studies in this area are required to elucidate which chemicals are being discharged and the sources of pollutants in order to reduce the risk of adverse effects upon the ecosystem.

PHA192 Effect Of Vitamin E-Defined Diet on Oxidative Stress Indicators in Channel Catfish (*Ictalurus punctatus*) Exposed to Benzo[a]pyrene and Dimethylbenz[a]anthracene. Chayasutabutr, A., Black, M.C.*, The University of Georgia, Athens, GA. Polycyclic aromatic hydrocarbons (PAHs), such as 7,12-dimethylbenz[a]anthracene (DMBA) and benzo[a]pyrene (BaP), are well known procarcinogens. Fish tissues, especially liver, have the ability to biotransform procarcinogens to DNA-reactive products. Once biotransformed, PAHs may generate high levels of reactive oxygen species (ROS), such as $\cdot O_2^-$, H_2O_2, and $\cdot OH$. These ROS can cause damage to DNA, including strand breakage and formation of oxidized bases. Antioxidant enzymes, including superoxide dismutase (SOD) and catalase, are also induced by conditions that increase the formation of ROS. Many antioxidant compounds, including vitamin E, can inhibit the formation of ROS. In this experiment the effects of dietary vitamin E on expression of several indicators of oxidative stress will be studied in channel catfish (*Ictalurus punctatus*). DNA damage, SOD and catalase activities were measured in catfish pretreated for 6 months with Vitamin E sufficient or deficient diets and subsequently exposed to BaP and DMBA. The hypothesis is that following exposure to DMBA or BaP, fish fed a vitamin E deficient diet will have increased DNA damage and higher levels of SOD and catalase activities than fish fed a vitamin E sufficient diet. Results are expected to confirm the protective effects of Vitamin E on oxidative stress.

PHA193 Biomarkers of Oxidative Stress in the Polychaete *Eurythoe complanata* (Amphinomidae) Under Short Term Copper Exposure. Nusetti, O.[1], [1]Universidad de Oriente, Escuela de Ciencias, Cumaná. Venezuela; Esclapés, M.M.*, PDVSA-INTEVEP, Ecology and Environment Management. P.O. Box 76343. Caracas 1070-A. Venezuela; Salazar, G. Instituto Universitario Tecnológico Sucre. Cumaná. Venezuela; Nusseti, S[1]. and Pulio, S[1]. There is ongoing research in our laboratory evoked to assess the relationship between heavy metal contamination and oxidative stress in benthic organisms from eastern littoral regions of Venezuela. Herein, we report the results of a study on antiperoxidant enzymes (glutathione reductase, glutathione peroxidase, catalase: potencial biomarkers of hydroperoxide detoxication) and microsomal malondialdehyde (MDA) levels (biomarker of oxidative stress) in the carcass tissue of the polychaete *Eurythoe complanata*, after 7 days exposure to 0.39 mg/L nominal concentration of Cu^{+2} (30% LC_{50}, from $CuSO_4.5H_2O$). The activity of glutathione reductase was significantly inhibed by the exposure (t-test = 12.6, $p<0.001$, N=6), whereas glutathione peroxidase and catalase were not affected (t-test < 1.120, $p>0.1$, N=6). The microsomal content of MDA increased significantly after 6 hours of exposure to copper, related to control values (2.354 ± 0.210 and 0.958 ± 0.076 nmol MDA/mg protein, respectively, N=3). At the end of the exposure time (168 h), concentration increased about ten fold (9.773 ± 0.722 nmol MDA/mg protein, N=3). The inhibition of glutathione reductase by copper exposure should decrease the cellular production of reduced glutathione. Consequently, the protective role of this molecule should be impaired, as reflected by the tissue accumulation of malondialdehyde. These observations suggest that the activity of glutathione reductase and the tissue levels of malondialdehyde, may be suitable biomarkers for screening acute chemical toxicity inducing oxidative stress in benthic organisms.

PHA194 Antioxidant Defenses in the Mangrove Mussel *Mytella Guayanensis*. Torres, M.A.; Tribess T.B.; Testa, C.P.; Pedrosa, R.C.*; Wilhelm Filho, D. Universidade Federal de Santa Catarina, Brazil. The digestive gland of forty five individuals of the mangrove mussel *Mytella guayanensis* collected in two polluted sites (Rio Sertao n=20, and Rio Itacorubi, n=20), and one nonpolluted site (Ratones, n=l5) were analyzed for different antioxidant defenses (AD). The cellular damage measured as TBARS levels were significantly enhanced ($P<0,05$) in the two polluted sites (Rio Sertao=1 86,5±26,6 nmoles g^{-1} and Rio Itacorubi 299,4±32,4 nmoles g^{-1}) compared to controls (132,1±38,6 nmoles g^{-1}). With the exception of the reduced form of glutathione (GSH=O,99±0,0,17 for Rio Sertao; 1,17±0,06 for Rio Itacorobi, and 1,19±0,O5 mM, for Ratones) and the enzyme superoxide dismutase (315,7±45,4 e 289,1±49,3 e 318,0±51, 6 U SOD g^{-1}, respectively), the other AD were significantly induced in polluted sites comparing with control site. The catalase activity (365,1±10,8 and 436,9±49,3 µmol $min^{-1} g^{-1}$, and 22,9±3,6 µmol min-1 g^{-1}; respectively) increased approximately one order of magnitude compared to controls. Similar enhancement were observed in the activities of glutathione reductase (14,57±2,23 e 7,70±1,13 mmol $min^{-1}g^{-1}$, and 0,48±0,13 mmol min^{-1} g^{-1} respectively). The activity of glutathione peroxidase increased 3-4 fold (158,1± 27,3 and 120,9±20,4 nmol $min^{-1} g^{-1}$), compared to Ratones (37,1±8,6 µmol min^{-1} g^{-1}), while the activities of glutathione S-transferase were significantly higher in the two polluted sites (24,1±1,5 and 16,8±1,3 µmol $min^{-1} g^{-1}$) compared to controls (10,4±1,4 µmol min^{-1} g^{-1}). The results indicate that *M. guayanensis* is routinely exposed to an oxidative stress in the polluted sites, as reflected in the higher cellular damage and the induction of most of the antioxidant enzymes. As a filter-feeding organism that favours the bioaccumulation of xenobiotics, the mangrove mussel is an excellent biomarker for biomonitoring studies.

PHA195 Glutathione Response of Estuarine Plants to Toxic Sediments. Lytle, J.S.* and Lytle,T.F., Institute of Marine Sciences, Gulf Coast Research Laboratory, The University of Southern Mississippi, Ocean Springs, MS 39564 Glutathione is widely distributed in plant cells and has been implicated in the adaptation of plants to environmental and chemical stressors. Glutathione acts as an antioxidant to protect labile macromolecules against attack by free radicals and hydrogen peroxide which are formed as a result of oxidative stress. Little in known regarding the sensitivity or adaptability of estuarine macrophytes to environmental toxins in coastal estuarine sediments. Though, under field conditions a number of environmental conditions may cause oxidative stress, this field study was designed to measure and compare glutathione responses from estuarine plants collected from sites whose major pollutant impact was known. A suite of estuarine plants were collected from four sites known to be impacted by certain xenobiotic compounds and from one site thought to be environmentally pristine. Glutathione content was compared among species in the same site to those species from the pristine site. Glutathione concentrations in *Juncus roemerianus* collected from a boat harbor contained levels as high as 1.6 μmoles glutathione/g dry wt as compared to 0.6 μmoles glutathione/g dry wt collected at the reference site. At those same sites, *Spartina alterniflora* had levels of glutathione elevated by 1.6X and *Scirpus robustus* by 4X that of the reference site. On the other hand, *Sesbania vesicaria* collected at a site containing methyl mercury (0.4 ppm in surface sediment) contained glutathione levels that were significantly diminished compared to the reference site. Glutathione responses varied among plant species at the same site as well as among sites for each individual species.

PHA196 ***In situ*** **monitoring of urban wastewater using transplanted mussels** ***Perna perna*** **in North Bay, Florianopolis, Brazil.** Bainy, A.C.D.*, Almeida, E.A., Medeiros, I.D. UFSC, Florianopolis, SC, Brazil. There is a crescent number of studies proposing the use of mussels as sentinel organisms in monitoring programs, since they are sessile, filter-feeders and possess good tolerance to adverse environmental conditions. However there is a controversy concerning the use of biomarkers in these organisms to express the impact of pollutants, because they are apparently less responsive to pollutants than other organisms. The purpose of this study was to analyze some biochemical responses in the digestive gland (DG) of mussels transplanted from a "reference" to a "polluted" site. Farmed mussels *Perna perna* (4-6 cm) were collected at Ilha de Ratones Grande ("Reference" site), caged individually and transplanted to a site that periodically receives the impact of urban wastewater discharges ("Polluted" site). A group was kept at the reference site in order to compare the results. After 30, 60 and 90 days, mussels from both sites (n=10 for each site) were killed and the DG was isolated and immersed in liquid nitrogen. A fragment of the DG was used to measure total glutathione (total GSH) content. The remaining portion of the DG was homogenized and centrifuged in order to obtain the cytosolic fraction in which the activity of superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) and glutathione transferase (GST) was determined according to conventional procedures. No significant changes in the activity of SOD, CAT, GST, as well as in the levels of total GSH were observed in the mussels kept at the polluted site for 30, 60 and 90 days, when compared to the reference group. However, lower GPx activity was observed in the mussels from the polluted site in both 30 and 60 days. These results show that the urban wastewater in North Bay is affecting the antioxidant defense system in the organisms exposed for shorter periods. Nevertheless, it is suggested the existence of a compensatory or adaptive mechanisms in the chronically exposed mussels (90 days). Additional experiments need to be done in order to quantify the levels of contaminants in these organisms, as well as in the water and the sediment at these sites.

PHA197 Differential Expression of the Multidrug Resistance P-Glycoprotein in Winter Flounder. Braud, M.C., University of Northern Iowa, Cedar Falls, IA; Laine, R.O.*, University of Florida, Whitney Laboratory, St. Augustine, FL. Many aquatic organisms are resistant to environmental pollutants and this resistance may be due to the function of their inherent multi-drug resistant glycoprotein extrusion pump (Pgp). This mechanism of multixenobiotic resistance is similar to the mechanism involved in multidrug resistance found in chemotherapy resistant human tumor cells in which the Pgp binds the cytotoxic drug and facilitates its efflux. If Pgp's are involved in drug extrusion or clearance, then Pgp levels should serve as an indicator or "biomarker" of exposure to chemical contaminants. Previously, two unique partial sequences coding for Pgp homologs in the genome of winter flounder were cloned by other investigators. We performed studies to demonstrate that the putative fish (winter flounder) Pgp gene is responsive to various toxins. This was determined with cultured primary hepatocytes in the laboratory using RT-PCR techniques. Primers were synthesized for the short regions that were previously sequenced and hepatocyte RNA was isolated from culture dishes previously incubated with the various toxins including polycyclic aromatic hydrocarbons, heavy metals, and model carcinogens. The use of these primers with RNA obtained from cells incubated in the presence and absence of the pollutant chemicals indicated an increased level of RNA for the two putative flounder Pgp clones compared with controls which used RNA from cells incubated in culture media only. Parallel studies investigating the expression of the gene products gave results that corresponded directly to the levels of RNA expression. Similar experiments also were performed using a transformed hepatocyte cell line. These preliminary results indicate, at the molecular level, the possibility of establishing this Pgp homolog of the human MDR gene as a diagnostic "biochemical marker" (ecosystem indicator).

PHA198 Peroxisome Proliferation in Aquatic Species: Does It Mean Anything? Haasch, M.L.*, Baier-Anderson, C. and Willis, S.M. Chesapeake Biological Laboratory, University of Maryland Center for Environmental Science, Solomons, MD, USA. Peroxisome proliferation consists of an increase in the volume and number of peroxisomes and the endoplasmic reticulum, and affects peroxisomal (β-oxidation), mitochondrial (carnitine acetyltransferase), cytosolic (epoxide hydrolase) and microsomal (Ω-oxidation; lauric acid hydroxylase (LA-OHase)) metabolism. Hepatocarcinogenesis has been noted in rats and mice after long-term PPA exposure. Environmental peroxisome proliferating agents (PPAs) include plasticizers, chlorinated solvents, bleached kraft pulp mill effluent, crude oils and petroleum products, lubricants, herbicides and insecticides. The pleiotropic response to PPAs is mediated by peroxisome proliferator-activated receptors (PPARs). Peroxisome presence was established in amphibians and fish in 1970 and 1974, respectively, but only recently has peroxisome proliferation been described in aquatic species (Fahimi and Cajaraville, 1995). Many invertebrates including insects, crustaceans, worms and mollusks undergo peroxisome proliferation and could be useful in biomonitoring of environmental contamination (Fahimi and Cajaraville, 1995). Amphibians exhibit species-specific responses to PPAs (*Rana esculenta* (responsive), *Xenopus laevis*; Ciolek and Dau•a, 1991), similar to the species-specificity of higher vertebrates. The presence of a PPAR has been described in Atlantic salmon (Ruyter et al., 1997) and zebrafish (P.M. Petkovich, pers. comm.). PPA exposure of rainbow trout produces no change in LA-OHase immunoreactive proteins (D.R. Buhler, pers. comm) similar to the response of Japanese medaka and zebrafish. Exposure of bluegill, channel catfish, fathead minnow and mummichog to PPAs produces increases (2-, 3-, 17- and 2-fold, respectively) in LA-OHase, PMP70, and iNOS immunoreactive proteins. These results suggest that several aquatic species could be susceptible to the same adverse consequences of PPA exposure as mammalian responsive species including developmental, reproductive, immunological and carcinogenic effects.

PHA199 Developments Towards an Immobilised ***Lux*****-based Biosensor.** Hall, J.M.*, Killham, K., University of Aberdeen, Aberdeen, Scotland. *Lux*-based biosensors are flexible, rapid and sensitive detectors of heavy metal and organic toxicity in water, soil and sediment samples. The development of on-line, batch and field toxicity tests based upon this technology is on-going. This study reports upon the progress to date of the development and potential of a portable field test kit based upon the use of immobilised *lux*-modified cells. A number of possible immobilisation strategies have been considered. One approach focused upon the immobilisation of microbial cells onto a range of surfaces with subsequent imaging using a Charge Coupled Device (CCD). A second approach has utilised the entrapment of microbial cells into gels. Alginate entrapped *lux*-based sensors were less sensitive to metals compared to the free cell bioassay. In contrast, the alginate immobilised sensor was equally sensitive to organic pollutants. The further development of this type of immobilised sensor may lie with applications that do not require the low detection levels of heavy metals. Immobilised sensors have been used successfully to report upon toxicity in digested sewage effluent samples. Current potential limitations of the immobilised system are the diffusion of oxygen (required for bioluminescence) and pollutants to the target sensor.

PHA200 DNA strand damage (Comet) assay and embryo development effects in grass shrimp, ***Palaeomonetes pugio*****, collected from sites contaminated with chromium, PCBs, mercury and toxaphene.** Lee, R.F.*, Kim, G.B., K. Maruya, Skidaway Institute of Oceanography, Savannah, GA; Steinert, S., Computer Sciences Corp., Marine Sciences Department, San Diego, CA. Grass shrimp are an important component of the estuarine food web along the coast of the eastern United States. We determined the amount of DNA strand damage and embryo development effects in grass shrimp embryos, which remain attached to the females during early development, from a number of contaminated sites in coastal Georgia and South Carolina. Site A is the site of a former chlor-alkali plant with inorganic mercury, methyl mercury and polychlorinated biphenyls at concentrations ranging from 10 to 100ppm. Site B is adjacent to a former toxaphene manufacturing site with toxaphene concentrations in the sediment ranging from 35 to 1800ppm. Site E is adjacent to a chrome plant where chromium in the sediment ranges from 2000 to 20,000ppm. DNA strand breaks, commonly known as the single-cell gelelectrophoresis method or the Comet assay, showed extensive DNA breakage(quantified as comet length or tail moment) in embryo cells from the most contaminated sites with significantly less damage as samples along a concentration gradient were examined. Arrested embryo development, poor hatching success and fewer embryos per female were found at the most contaminated sites compared with clean reference sites. Correlations were found between contaminant concentrations, DNA strand damage and embryo development effects.

PHA201 5-bromo-2'-deoxyuridine Labeling of Hepatocytes as an Indicator of Cellular Proliferation in the Japanese Medaka fish (*Oryzias latipes*). Brennan, L.M.*, GEO-CENTERS, INC. @USACEHR, Fort Detrick, MD; Boncavage-Hennessey, E.M.., GEO-CENTERS, INC. @ Colorado State University, Fort Collins, CO; Toussaint, M.W., GEO-CENTERS, INC. @ USACEHR, Fort Detrick, MD; Wolfe, M.J., Experimental Pathology Laboratories, Inc., Herndon, VA; Gardner, H.S., USACEHR, Fort Detrick, MD. The Japanese medaka (*Oryzias latipes*) has been used extensively as a nonmammalian species for chemical carcinogenicity testing. Mammalian studies have demonstrated that increased cellular proliferation is often an early and necessary step in the multistage process of chemically-induced hepatocellular carcinogenesis. Early increases in hepatocellular proliferation were examined and compared in the medaka after exposure to a suspect nongenotoxic carcinogen, chloroform, and after exposure to the known genotoxic carcinogen, diethylnitrosamine (DEN). Proliferation was assessed by exposing the medaka to the thymidine analog 5-bromo-2'-deoxyuridine (BrdU) in the aquarium water for 72 hours, sacrificing the fish for immunohistochemical processing, and then measuring the S-phase labeling area index. Fourteen-day old medaka were exposed to 0.015, 0.15, or 1.5 mg/L chloroform in the water under continuous flow-through conditions. Subgroups of fish sacrificed after 4 and 20 days of chloroform exposure did not show increased hepatocellular proliferation as compared to controls. Fish from this experiment that remained on study after 6 months of continuous chemical exposure also did not have an increase in liver neoplasms as compared to controls. Fourteen-day old medaka exposed for 48 hrs to a single concentration of 10 or 100 mg/L DEN in the water had significant increases in hepatocellular proliferation at 7 days post-DEN exposure; 100mg/L fish had increased proliferation at 14 days, as well. At the 6-month sacrifice point, fish that had received high-dose DEN also had significantly more liver neoplasms than control fish. Preliminary results indicate the usefulness of BrdU-labeling of S-phase hepatocytes in the medaka as an indicator of the potential for a compound to induce cellular proliferation in a target organ.

PHA202 Randomly Amplified Polymorphic DNA Markers as Reliable Biomarkers in Evaluation of Genetic Integrity in Small Mammals from a Hazardous Waste Site. Sternberg, D.C.*, Wright State University, Dayton, OH, Sheffield, S.R., Clemson University, Pendleton, SC, Krane, D.E., Wright State University, Dayton, OH, Lochmiller, R.L., Oklahoma State University, Stillwater, OK, Burton, G.A., Jr., Wright State University, Dayton, OH. Randomly amplified polymorphic DNA (RAPD) markers have proven to be a promising tool for the evaluation of genetic variability and integrity in natural populations. Recently, this technique has been successfully used in the genetic evaluation of populations of several species inhabiting contaminated aquatic sites in Ohio. In the current study, genetic profiles of three small mammal species (*Sigmodon hispidus, Reithrodontomys fulvescens, Peromyscus maniculatus*) from a hazardous waste site and uncontaminated reference sites were evaluated and compared. The hazardous waste site is a former oil refinery in south-central Oklahoma that operated from about 1920 into the 1980's and currently is on the EPA National Priorities List (Superfund). Contaminants at the site include a wide variety of toxic metals and organics, including lead, chromium, arsenic, benzene, toluene, phenols, and PAHs. Preliminary results using two independent rapid PCR primers indicate that there is decreased genetic diversity in each species of mammal from the hazardous waste site when compared to reference site animals. Currently, we are in the process of analyzing a much larger sample size of each species in order to attain better statistical validation of this finding. These results provide further evidence that RAPD is a tool capable of evaluating the genetic integrity of a population in areas impacted by chemical contamination.

PHA203 The impacts of dissolved copper on rainbow trout (*Oncorhynchus mykiss*) in a Sierra Nevada stream. Dethloff, G.M.*, University of California, Davis, CA; Maier, K.J., University of Memphis, Memphis, TN. Rainbow trout (*Oncorhynchus mykiss*) were sampled from two creeks in the western Sierra Nevada, Plumas County, CA. Dissolved copper (Cu) concentrations are elevated in one creek after receiving water from a small tributary that flows through a 40-hectare Cu tailings pile from a now-sealed mine. Rainbow trout were collected at two reference sites and at two sites that receive Cu effluent. Hepatic Cu concentrations were significantly elevated in trout at both sites downstream of the Cu source. Depressions in hematocrit, leukocrit, and the percentage of lymphocytes in the blood occurred in trout at the Cu-contaminated sites. Measurements of the percentage of monocytes in the blood and respiratory burst activity of trout were affected by gender and age, respectively. Condition factor, plasma acetylcholinesterase, percentage of neutrophils in the blood, muscle glycogen and muscle protein were not affected by dissolved Cu concentrations. The data from this study support the use of immune system parameters to assess alterations in salmonids affected by Cu contamination and illustrate the variability in physiological responses of wild fish caused by demographic features such as age and gender.

PHA204 *Lux*-based Biosensors - Reporting on Metal Bioavailability in Synthetic Soil Solutions. Hall, J.M.*, Sarin, C., Rattray, E.A.S., Killham, K. Cresser, M.S., Cotter-Howells, J. C., University of Aberdeen, Aberdeen, Scotland. *Lux*-based biosensors have been found to be flexible, rapid and sensitive detectors of heavy metal toxicity in water, soil and sediment samples. This study investigated the basis of the bioluminescence response to metal toxicity. Standardised freeze-dried batches of *E. coli* were exposed to a range of metals (e.g. Pb, Cu, Zn, Hg) prepared in synthetic soil solution. Four different soil solution ionic strengths were created by the addition of varying amounts of K_2SO_4. EC_{50} values were calculated from the decline in bioluminescence, and free ion concentration in the test solutions was predicted using the chemical speciation model, GEOCHEM-PC. In general, as soil solution ionic strength increased, EC_{50} values also increased indicating reduced toxicity based on total metal concentrations. However, EC_{50} values were similar when expressed as free ion concentration. The *lux*-based biosensor appeared to respond to free ion concentration in most cases. This work in combination with previous findings, suggests that *lux*-based biosensors report on toxicity as free metal ion concentrations or on the most toxic metal complex present, and may be used to assess the bioavailability of metals in environmental samples.

PHA205 Effect of Dietary Exposure of the Tunicate *Amouricium sp.* on Hepatic Expression of CYP1A and GSTÕs in the Pinfish, *Lagodon rhomboides*. DeBusk, B.C., Slattery, M, and Schlenk, D. Environmental Toxicology Research Program/RIPS, Department of Pharmacology, School of Pharmacy, University of Mississippi, University, MS. The effects of diet and other non-anthropogenic effects on biomarker expression in various organisms have been largely ignored. The goal of this study was to examine the effects of dietary exposure to a tunicate (*Amouricium sp.*) extract on biomarker expression in the tunicate-consuming pinfish (*Lagodon rhomboides*). Tunicates and juvenile pinfish (approximately 8 cm in length) were collected from shallow coastal waters of Florida. The fish were housed for 9 months in a recirculating seawater system. Fish were then isolated individually into 10L aquaria and fed krill food cubes containing either vehicle or 100 mg of tunicate extract per day for two days and 200 mg of tunicate extract per day for three additional days. At the end of the fifth day, hepatic microsomes from individuals were probed for expression of CYP1A by Western blotting using an anti-CYP1A peptide monoclonal antibody. Cytosolic fractions were examined for expression of three GST families, a, m, and p, by Western blotting using polyclonal antibodies raised against mammalian GSTÕs (provided by Dr. Evan Gallagher). GST activity was examined using a microplate assay of CDNB conjugation. Hepatic CYP1A in pinfish was induced 4-fold by the extract. Expression of a 20 kD protein recognized by antibodies raised to GST α was unchanged. Expression of a 20 kD protein recognized by antibodies raised to GSTÕs mu and pi tended to decrease in treated animals, though not significantly. CDNB conjugation activity was unchanged by treatment. These results indicate that dietary exposure to natural organic compounds can significantly alter detoxification (biomarker) responses in pinfish.

PHA206 White Clover as a Sentinel Bioindicator for Ambient Ozone: Comparison of Detection of Effects by Relative Response of Clones or Ethylenediurea EDU) Applications. Manning, W.J.*, Johnson, M.S., and Frenkel, M.E. University of Massachusetts, Amherst, MA. Ladino white clover *(Trifolium repens* L. 'Regal') can be used to estimate air quality for ambient ozone. Clone NC-S is sensitive to ozone, while NC-R is tolerant. The proportional response of the two clones (NC-S/NC-R response ratio), expressed as changes in shoot biomass after exposure to ambient ozone, is a quantitative assessment. NC-R is assumed to be not affected by ozone while NC-S incurs injury expressed as growth reduction. If the ratio of NC-S/NC-R is less than one, then it is assumed that ozone concentrations caused plant injury. One way to test this relationship is to apply the ozone-injury suppressant chemical ethylenediurea (EDU) to half of the NC-S and NC-R plants to determine whether or not results would be the same and whether NC-R does respond to ozone, thus changing the value of the ratio. It would also determine whether NC-R is necessary and whether or not the NC-S response/NC-S response plus EDU is a more accurate plant response to ozone. We grew potted clover plants of both clones in the field during the growing seasons of 1994-1997. Half the plants were sprayed with EDU weekly. Shoot biomass was harvested every 28 days and expressed as dry weights. Depending on the weather and ozone concentrations in a given year, EDU applications could be used to verify that differences in NC-S/NC-R ratios were caused by ambient ozone. In some cases, however, EDU also increased the biomass response of NC-R, indicating that it was also affected by ozone. NC-S response/NC-S plus EDU is probably a better indicator for ozone than NC-S/NC-R.

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