SYSTEMATICS OF *CAENODELPHAX* FENNAH (HEMIPTERA: FULGOROIDEA: DELPHACIDAE) AND DESCRIPTION OF THE NEW GENUS *FLAVOCLYPEUS*

by

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A thesis submitted to the Faculty of the University of Delaware in partial fulfillment of the requirements for the degree of Master of Science in Entomology

Winter 2013

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ACKNOWLEDGMENTS

First and foremost, I would like to thank my advisor, Charles Bartlett, whose guidance was invaluable to me in every aspect of this project. I am also indebted to my graduate committee members, Deborah Delaney and Carl Schmidt, for their support. Additionally, my colleague Kathryn Weglarz provided vital collaboration and counsel that greatly expedited my progress on this project.

I am grateful to Julie Urban of North Carolina State University, Jason Cryan of the New York State Museum, Steve Wilson of Central Missouri State University, Andrew Hamilton of the Canadian National Museum, and Brewster Kingham of the Delaware Biotechnology Institute for their help on this project. For the Ioan of specimens, I gratefully acknowledge Chris Dietrich of the Illinois Natural History Survey, Toby Schuh of the American Museum of Natural History, Stuart McKamey and Michele Touchet of the Smithsonian Museum of Natural History, Bob Blinn of the North Carolina State University, Shepherd Myers of the Bishop Museum in Hawaii, Michael Sharkey of the University of Kentucky, Lois O'Brien, Mick Webb and Max Barclay of the British Museum, Richard Hoebeke of Cornell University Department of Entomology, and Jérôme Constant of the Institut Royal des Sciences Naturelles de Belgique. I am indebted to Tom Ho for assistance with laboratory work, Lawrence Barringer and Anthony Deczynski for collecting specimens, and Kimberley Shropshire for assistance in photography. I thank Andrew Short of the University of Kansas for giving me the opportunity to travel to Costa Rica to collect specimens with his team.

For his invaluable assistance with phylogenetics, as well as his tireless encouragement, I gratefully recognize Cyrus Kasaaian, who did not allow his strong aversion to insects to get in the way of helping me. I would like to acknowledge the other students in the ENWC department, most notably the bee folk and Bill Cissel, who helped to make my graduate experience a truly memorable time of personal and academic growth. Lastly but certainly not least, I am grateful to my parents who have always supported me in every endeavor.

This research was made possible through the USDA National Institute of Food and Agriculture Grant 2009-55605-05006 and the University of Delaware Department of Entomology and Wildlife Ecology.

NOMENCLATURAL NOTE

This work is considered unpublished for the purposes of zoological nomenclature (Art. 8.3, ICZN 4^{th} ed., 1999).

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ABSTRACT

Delphacid planthoppers (Hemiptera: Fulgoromorpha: Delphacidae) are of worldwide economic interest as crop pests and vectors of plant diseases. Despite their importance, much of their evolutionary history remains poorly understood and many genera within Delphacidae need revision. *Delphacodes* Fieber, 1866 once included more than 136 species, including many New World species, but was redescribed with a more limited definition, reducing it to only 10 western Palearctic species. This left the majority of *Delphacodes* species in need of reassignment to other genera.

Hamilton (2002) hypothesized that 10 New World *Delphacodes* species belong to *Caenodelphax* Fennah, 1965. This project undertook an investigation of Hamilton's hypothesis by examining a subset of 13 *Delphacodes* and 4 *Caenodelphax* species with reference to morphological phylogenetic analyses to determine their evolutionary relationships. Phylogenetic analyses using maximum parsimony did not support Hamilton's hypothesis, and instead suggested that eight ingroup species belong in a separate, new genus. *Caenodelphax* is redescribed here as a monotypic taxon; eight species are transferred to the new genus, *Flavoclypeus*, and two species are synonymized.

Chapter 1

GENERIC REVISION OF CAENODELPHAX

1.1 Introduction

Planthoppers belong to the order Hemiptera, consisting of the true bugs. As seen in other hemipterans, planthoppers are paurometabolous (having incomplete metamorphosis, lacking a pupal stage, with adults and immatures occurring in the same habitat) and have piercing-sucking mouthparts, which they use to extract juices (mostly phloem) from plants. Planthoppers comprise the infraorder Fulgoromorpha, with the single extant superfamily Fulgoroidea, which along with their sister group, the infraorder Cicadomorpha, makes up the suborder Auchenorrhyncha. Auchenorrhynchans are marked by having uniform textured wings, jumping hind legs, and a tymbal or sound-producing organ. Fulgoromorphs, in turn, are characterized by their antennae arising below the compound eyes and bearing carinae on the head and thorax. Planthoppers are found in many biomes, including deserts, tropical rainforests, grasslands, and the arctic tundra (Wilson et al. 1994). Of the twenty recognized families that make up Fulgoromorpha, the largest and most well-known is the Delphacidae, with more than 2100 known species (Wilson 2005).

1.1.1 Background of the family Delphacidae

The family Delphacidae is considered one of the most primitive, basal families within the Fulgoroidea, along with the closely-related Cixiidae, and likely arose prior to the Cretaceous period (145-70 million years ago). Delphacid planthoppers are most easily distinguished from other families by the presence of a distinctive moveable spur, or calcar, at the apex of each hind tibia (Figure 1). The calcar is found in both adults and immatures, frequently bears teeth, and can be used as a diagnostic feature in identification. Additionally, delphacids have a row of apical spines on the second hind tarsomere, pincer-like parameres, and a sword-shaped ovipositor. The male genitalia are highly species-specific; dissection is often necessary to identify specimens to species level. Females of many species cannot be positively identified unless they are found in association with males. Delphacids are typically small (roughly 2-4 millimeters in length) and cryptic in both color and habit. Brachyptery, or the condition of having short wings that do not completely cover the abdomen, occurs in many species, although macroptery (long-wingedness) is more common, and both forms occur within the same populations (Figure 2). Macropters display much better dispersal and long-distance flight capabilities than brachypters, although brachypters have the advantages of earlier reproduction and higher fecundity.



Figure 1. Hind leg, including calcar, of *Delphacodes recurvata*.

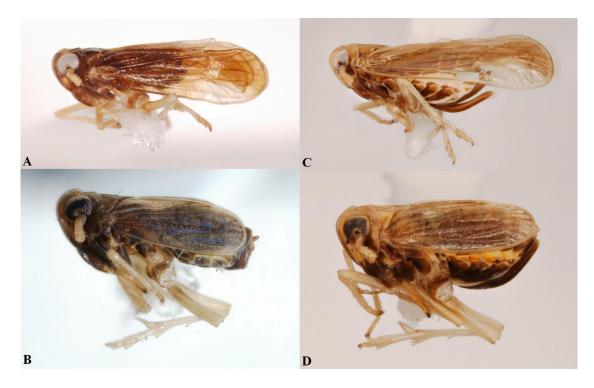


Figure 2. Variation within *Caenodelphax teapae* by sex and wing morph. A. Male macropter, B. Male brachypter, C. Female macropter, D. Female brachypter.

Among the Delphacidae are many important crop pests found worldwide, most notably *Nilaparvata lugens*, the brown planthopper, *Perkinsiella saccharicida*, the sugarcane planthopper, and *Peregrinus maidis*, the corn planthopper (Wilson 2005). As a group, delphacids tend to be highly host-specific, with a majority feeding on phloem sap in monocots, usually grasses and sedges in the families Poaceae, Cyperaceae, and Juncaceae, although oceanic island species are more likely to be dicot feeders. Like many other phytophagous species, their nutritional requirements are met via assistance from beneficial endosymbionts passed from generation to generation. These symbiotes provide nutrients that are otherwise unavailable in the host plant such as cysteine, methionine, and tryptophan. In addition to mechanical damage ("hopperburn") caused by feeding on vascular tissues of plants and by cutting slits into plants for oviposition, delphacids are vectors for more than thirty known plant viruses and at least one phytoplasma (Wilson 2005). Among the economically important crops they attack are sugar cane, maize, rice, wheat, barley, and oats. In many cases, modern agricultural practices have led to surges in planthopper populations and greater difficulty in control, often due to declining natural predator populations from pesticide application. Wolf spiders are one such natural enemy (Cronin et al. 2004; Denno et al. 2004), capable of keeping planthopper populations in check when their numbers are not reduced by agrochemical practices (Liu et al. 2001).

In spite of delphacids' notoriety as a group containing many pests, much of their taxonomy is shrouded in uncertainty; many genera require systematic revision. The overarching aim of systematics is to clarify phylogenetic relationships among living things, a process that involves both molecular and morphological research as well as phylogenetic analysis. Relationships are depicted in phylogenetic trees that reveal evolutionary history. Systematists strive to uncover monophyletic groups, that is, clades which consist of a single common ancestor and all the descendants of that ancestor. In contrast to monophyly, a group may be *paraphyletic* if it consists of an ancestor species and only some of its descendants or *polyphyletic* if it consists of unrelated taxa mistakenly grouped together. Among the delphacids are many genera known or suspected to be polyphyletic.

The largest subfamily within the delphacids is Delphacinae, a monophyletic grouping that consists of 3 tribes: Saccharosydnini, Tropidocephalini, and Delphacini.

Delphacinae's monophyly is supported by synapomorphies such as the thin-walled central portion of the aedeagus. Delphacini is in turn the largest tribe within Delphacinae, containing approximately 1600 species (about 75% of known delphacid species), and is also believed to be a monophyletic grouping (Urban et al. 2010). The Delphacini are characterized by a suspensorium (internal brace for the aedeagus) with a distinct Y, O, or U shape (Asche 1985, 1990) and also by the presence of endosymbionts H and f and the absence of symbiont x, recently suggested to be the Betaproteobacterium *Vidania* (Urban and Cryan 2012).

1.1.2 History of Delphacodes

Delphacodes is a large genus within the tribe Delphacini that is widely regarded as a "junk genus" due to its polyphyletic status. It was originally established by F.X. Fieber in 1866 as a subgenus of *Delphax* containing only 10 species, but in 1904 it was raised to genus level by G.W. Kirkaldy. Kirkaldy (1904: 177) designated a female lectotype of *Delphacodes mulsanti* as the type specimen for the genus. The lectotype's sex and imperfect condition contributed to the uncertainty and imprecision in the genus's definition, leading to a broad, loose interpretation of *Delphacodes* and consequently leading to the inclusion of many unrelated species under this grouping. At one time, *Delphacodes* included 136 New World species, in addition to numerous Old World species. In 1963, W. Wagner redefined it more narrowly, limiting it to only 10 western Palearctic species. This left many other species, including all of the New World taxa, *incertae sedis* and in need of reassignment to other genera. Recent work by Urban et al. (2010) has unequivocally demonstrated the polyphyly of *Delphacodes*; their phylogenetic tree compiled using mixed model Bayesian analysis with morphology and four genes reveals *Delphacodes* species occurring in multiple branches. Further complicating the *Delphacodes* problem is the scarcity of comprehensive species keys and the fact that many species are difficult to find.

1.1.3 History of *Caenodelphax*

In contrast to *Delphacodes*, the genus *Caenodelphax* is small, heretofore consisting of only 4 species, but it too has had a complicated taxonomic history. R. G. Fennah first described *Caenodelphax* in 1965. At that time, it included only two species, both Neotropical: *C. teapae* and *C. philyra*. The type species, *C. teapae*, was first described as *Liburnia teapae* by Fowler, 1905 based on several specimens from Teapa, Mexico, and then subsequently reassigned to *Megamelus* by D. Crawford (1914). Later, it was transferred to *Delphacodes* by G.N. Wolcott (1923) before Fennah finally ascribed it to *Caenodelphax* in 1965. *Caenodelphax philyra* was first described as *Delphacodes philyra* by Fennah in 1959 before he moved it to *Caenodelphax* along with *C. teapae* in 1965.

The genus doubled in size when K. G. A. Hamilton (2002) and P. Bouchard and colleagues (2002) added two more species to *Caenodelphax*. These species were initially described as *Delphacodes nigriscutellata* and *D. atridorsum* by R.H. Beamer in 1947. Bouchard et al. (2002) transferred the former and Hamilton (2002) transferred the latter to *Caenodelphax*. Both *C. nigriscutellata* and *C. atridorsum* are Nearctic, but because they are not Neotropical, these taxonomic acts changed the definition of *Caenodelphax* from a strictly Neotropical genus to a more widely distributed New World genus. At the same time, Hamilton indicated that an additional 10 Nearctic species currently in *Delphacodes* belong in *Caenodelphax*. Although he did not denote which taxa should be moved, he specified that they occur in eastern and central North America and gave several key features by which to identify them.

The key features Hamilton identified as offering support for the inclusion of certain *Delphacodes* species in *Caenodelphax* include similarities in color and structure. For example, a marked contrast between the pale antennae and dark frons occurs in most species. Hamilton (2002) observed that delphacid color patterns tend to be conserved within genera and indicated that the pale antennae-dark frons pattern may serve as a useful diagnostic feature for *Caenodelphax*. He noted that other key diagnostic features of *Caenodelphax* are a narrow crown, black dorsum, and small, knife-shaped calcar. Other delphacid species with narrow crowns have large, leaf-shaped calcars, and other species with small, knife-shaped calcars have wide crowns (Hamilton 2002).

1.2 Project Goals

This project undertook a revision of *Caenodelphax* to test Hamilton's hypothesis of which characters distinguish this genus and which species it comprises. Seventeen species, including the four pre-established *Caenodelphax* species and a segregate of 13 species from the polyphyletic *Delphacodes*, were examined on a

morphological basis to determine the characters that differentiate them and the characters they have in common. A phylogenetic analysis was completed to establish a new hypothesis for the evolutionary relationships among the taxa, and a new generic key was written to reflect this hypothesis. Uniform descriptions and photos for all included species were completed and the website for the overarching project on delphacids of North America was updated to include this taxonomic revision. The first chapter presents the materials and methods and the results of the phylogenetic analyses; the second presents the systematic treatment of the examined species.

1.3 Materials and Methods

Seventeen species of delphacids were examined for taxonomic revision (Table

1). In total, 887 specimens were examined for this study.

Species	Author, Year	Type Location
Caenodelphax atridorsum	(Beamer, 1947)	SEMC
Caenodelphax nigriscutellata	(Beamer, 1947)	SEMC
Caenodelphax philyra	(Fennah, 1959)	BMNH
Caenodelphax teapae	(Fowler, 1905)	BMNH
Delphacodes adunca	Beamer, 1948	SEMC
Delphacodes andromeda	(Van Duzee, 1907)	CASC
Delphacodes aterrima	Muir, 1926	BPBM
Delphacodes balli	Muir and Giffard, 1924	CASC
Delphacodes incurva	Beamer, 1948	SEMC
Delphacodes latidens	Beamer, 1948	SEMC
Delphacodes livida	Beamer, 1948	SEMC
Delphacodes nigrifacies	Muir, 1918	AMNH
Delphacodes nitens	Muir and Giffard, 1924	BPBM
Delphacodes recurvata	Beamer, 1948	SEMC
Delphacodes shermani	(Metcalf, 1923)	NCSU
Delphacodes sucinea	Beamer, 1948	SEMC

Table 1. Species considered for inclusion in *Caenodelphax* with authors and locations of types.

1.3.1 Specimen acquisition and included material

The acquisition of specimens representing the ingroup taxa was the first step in this combined phylogenetic analysis. Most of the 17 species hypothesized to be part of *Caenodelphax* were represented in the University of Delaware collection; others were acquired on loan from other institutions (Table 2). Samples of *D. andromeda* were collected via sweep-netting in Maryland. Additionally, I took part in a multi-institution field expedition to Costa Rica in July 2011 to procure supplementary specimens of the tropical taxa, namely *C. teapae* and *D. aterrima*. These specimens were acquired via sweep-netting along paths at Tapantí National Park and using an aspirator at light traps at Kiri Lodge on the outskirts of the park. Collected specimens were retained in alcohol for molecular analysis or glued to points on the right side of the thorax and placed on insect pins and labeled with collection data.

Label data were recorded for all included specimens. For type material, label data were quoted verbatim using "/" to indicate a line break and "//" to indicate a new label and with supplemental information given in brackets (e.g., [handwritten, folded red label]). For other material examined, label data were rewritten to maintain consistency in pattern, beginning with the country, state or province, and more specific locality, followed by the collection date, collector, and lastly the number and sex of specimens and the depository where the specimens are located, given in parentheses. Additional information such as elevation, GPS coordinates, host plant, and collection

method were included, if given, in the same order as seen on the label data.

Abbreviations in label data were expanded for clarity, except in cases in which the full

expanded name was not immediately clear.

The distribution of all taxa was inferred from the available specimens;

additional localities reported in the literature record were also included. Collections

from which specimens of described taxa were examined are abbreviated following

Arnett et al. 1993 (Table 2), with the addition of the Louisiana State Arthropod

Collection (LSAM) and the University of Kentucky Collection (UKYC).

Table 2. Collections that loaned specimens for this project.		
Codon	Collection	Number of specimens by sex
AMNH	Department of	1 \bigcirc , 1 \bigcirc <i>D. nigriscutellata</i>
	Entomology Collection,	$1 \bigcirc D$. incurva
	American Museum of	
	Natural History, New	
	York, NY	
BMNH	Department of	$1 \stackrel{\wedge}{O} C$. teapae
	Entomology, The Natural	$1 \stackrel{?}{\circ} C. philyra$
	History Museum, London,	
	United Kingdom	
BYUC	Monte L. Bean Life	$5^{\uparrow}, 2^{\bigcirc}, 1$ broken <i>C. teapae</i>
	Science Museum,	
	Brigham Young	
	University, Provo, UT	
CDAE	California State Collection	9Å, 2 ♀ <i>C. teapae</i>
	of Arthropods, Analysis	
	and Identification Unit,	
	California Department of	
	Food and Agriculture,	
	Sacramento, CA	
CUIC	Cornell University Insect	14 \bigcirc <i>C. teapae</i>
	Collection, Department of	
	Entomology, Cornell	
	University, Ithaca, NY	
ISNB	Collections Nationales	2 \bigcirc D. andromeda
	Belges d'Insectes et	

Table 2. Collections that loaned specimens for this project.

	d'Arachnides Institut	
	d'Arachnides, Institut	
	Royal des Sciences	
	Naturelles de Belgique,	
LDOD	Brussels, Belgium	
LBOB	Lois O'Brien Collection,	5 , 1 \bigcirc <i>D. nigrifacies</i>
	Green Valley, AZ,	
	associated with CASC	-
LSAM	Louisiana State Arthropod	$2 \stackrel{\bigcirc}{\downarrow} D$. andromeda
	Museum, Louisiana State	7♂, 3♀ <i>D. latidens</i>
	University, Baton Rouge,	
	LA	
SEMC	Snow Entomological	2 \eth , 1 \bigcirc <i>D. nigriscutellata</i>
	Museum, University of	103, 99 D. incurva
	Kansas, Lawrence, KS	
TAMU	Department of	1 $\mathcal{J}, 1 \mathcal{Q}$ <i>C. teapae</i>
	Entomology Insect	5, 17 , D . latidens
	Collection, Department of	$6^{\uparrow}, 9^{\bigcirc}$ D. nigrifacies
	Entomology, Texas A&M	8♂ D. nitens
	University, College	
	Station, TX	
UDCC	Department of	59 3 , 8 \bigcirc <i>C. teapae</i>
	Entomology and Wildlife	$1 \stackrel{?}{\partial} D. adunca$
	Ecology Collection,	70♂, 97 $\stackrel{\circ}{\downarrow}$ D. andromeda
	University of Delaware,	123, 40, 1 broken <i>D. latidens</i>
	Newark, DE	823, 789 D. nigrifacies
		3♂ D. nigriscutellata
		$173, 9$ $\stackrel{\frown}{\rightarrow}$ D. nitens
UKYC	Department of Biology	29°, 22 ° D. nigrifacies
	Collection, University of	
	Kentucky, Lexington, KY	
USNM	United States National	1233, 299, 2 broken <i>C. teapae</i>
	Entomological Collection,	23, 59 C. atridorsum
	Department of	$22^{\uparrow}, 2^{\bigcirc}, 1$ broken <i>D. adunca</i>
	Entomology, U.S.	103 D. andromeda
	National Museum of	7♂, 4♀ <i>D. latidens</i>
	Natural History,	10° , 7° D. nigrifacies
	Smithsonian Institution,	73, 39, 1 broken D. nitens
	Washington, DC	

1.3.2 Morphological analysis

The morphological portion of this project entailed examining the type specimens, if available, and other specimens for each of the ingroup taxa to review the current understanding of each species' definition. Each ingroup species was redefined in a consistent style. Illustrations of each species, including photos of dorsal and lateral views, the frons, and lateral and caudal views of the male genitalia were incorporated as needed. Morphological terminology follows Asche (1985), except "segment 10" is substituted for "anal tube" and "armature" is used in reference to the aedeagal brace on the diaphragm.

Male genitalia were dissected for description and identification. The abdomen was removed and cleared in potassium hydroxide (KOH) overnight, rinsed in water, and transferred to glycerol for observation. Dissected parts were retained with glycerin in microvials pinned with the specimens.

The type specimen serves as the standard bearer or *de facto* definition for the entire species, to which all other specimens should be compared. Similarly, the type species is designated as the standard bearer for the genus. *Caenodelphax teapae* is the type species for the genus *Caenodelphax*; this species defines the features of the genus. Morphological revision includes coloration as well as size, structure, and the presence or absence of physical features. A variety of male and female specimens from various localities were included in morphological analyses to account for potential geographic variation or sexual dimorphism.

All observations were made using a Wild Heerbrugg dissecting scope with 20x oculars and a 6-50x objective lens. All photographs and measurements were taken using a Nikon SMZ-1500 Digital Imaging Workstation with Nikon DS-U1 digital camera and NIS Elements Imaging software (Version 3.0); photos were compiled into plates using FastStone Image Viewer (Version 4.6). Reported measurements are averages in millimeters, with the number measured (n) specified; some measurements are expressed in the descriptions as ratios of length to width (l:w). Total body length was defined as the length from the tip of the vertex to the wing tip in macropters and from the tip of the vertex to the tip of the abdomen in brachypters; width was defined as the distance across the mesothorax between the tegulae. The length and width of antennal segments I and II were measured at the widest points. Frontal length was measured along the median carina from the vertex to the frontoclypeal suture; frontal width was measured across the lateral margins, between the antennae. Pronotal and mesonotal length were measured along their respective median carinae. Calcar length was defined as the distance from the articulation with the tibia to the apex of the calcar. In the event that a wing morph or sex was not observed, its omission was specified under the "Structure" heading.

The nomenclature of the included species was reviewed in order to ensure that all names are nomenclaturally valid, as stipulated by the International Code of Zoological Nomenclature (ICZN, 1999). The ICZN's guidelines concerning synonymizations were also applied in two cases of synonymy to determine which of the two names should take precedence.

Type specimens for each species, or other representative specimens if the types were not available, were compared with the accepted species definition to ensure that they correspond. When possible, the type specimen was photographed. With a revised understanding of the species definition, all the available specimens of the ingroup taxa were reviewed. The biology and host plant data from the literature and from the specimen labels were compiled, along with distribution information, so as to provide a complete, comprehensive species description for all ingroup taxa. Host plant scientific and common names are provided based on the USDA PLANTS database (USDA, NRCS 2010).

1.3.3 Molecular analysis

Mitochondrial DNA sequence data from the cytochrome oxidase I gene was extracted for all available ingroup species for phylogenetic analysis. Some species were excluded from molecular analysis because they were only represented by dry, pinned specimens; molecular work necessitates preservation in alcohol. Two species, *Caenodelphax philyra* and *Delphacodes xerosa*, ultimately determined to be junior synonyms of *C. teapae* and *D. nigrifacies*, respectively, were only known from the holotypes and consequently were not included in molecular analysis in order to avoid damage to the types.

The general outline of the molecular methods was as follows: extraction of total DNA from thoracic or leg tissue, amplification of mitochondrial DNA via PCR using AmpliTaq DNA polymerase, gel electrophoresis with ethidium bromide staining

to visualize the amplified gene, and sequencing. Using a precedent established by other delphacid taxonomists (Dijkstra et al. 2003), I used COI, a mitochondrial barcoding gene. This protein-coding gene is the main subunit of the cytochrome C oxidase complex. COI is a fast-evolving gene found in all eukaryotes, and its variability renders it appropriate for determining shallow splits in the phylogenetic tree, such as species within a genus.

Extraction of mitochondrial DNA entails three separate steps: cell lysis, purification, and elution. Cell lysis, or the process of breaking open cell membranes to allow DNA release, is completed using a cell lysis solution, Buffer ATL. The enzyme proteinase K must also be added to the solution to denature the proteins in the insect tissue so that they cannot break up the DNA. Ethanol is also added to precipitate the DNA and the buffer AL is used to split the DNA strands. The elimination of cellular debris is accomplished by placing the cellular components into a spin column for centrifugation, enabling the debris to fall through, and followed by washing twice with the buffers AW1 and AW2, leaving a purified sample. DNA elution involves using the elution buffer AE to pull the DNA into the collection tube. Polymerase chain reaction involves repeatedly heating and cooling to facilitate enzymatic replication. A Bio-Rad 96-well MyCycler[™] thermocycler was used for PCR. The oligonucleotide primers Pat and COI-RLR (Simon et al. 1994) and the heatstable DNA polymerase AmpliTaq were used to enable amplification; during amplification, the DNA generated is used as the template for replication. Initially, the primers Ron and Calvin (Simon et al. 1994) were used but failed to produce satisfactory amplification results; Pat similarly yielded poor amplification results, but its use was maintained due to lack of better known alternatives. Agarose gel electrophoresis was performed with ethidium bromide staining to visualize the amplified DNA using a Bio-Rad Power Pac 1000 power supply.

An ethanol precipitation procedure was performed to purify and concentrate the DNA. Sodium chloride was added to a solution of 100% ethanol and the PCR product to provide positive ions; the DNA and salts combined to form a precipitate that was collected by centrifuging, using a Thermo Scientific Sorvall Legend Micro 21 Microcentrifuge. The supernatant was discarded to leave a pellet of crude DNA which was dried and resuspended in 25 microliters of nuclease-free, double-distilled water in preparation for sequencing.

Analysis at the Delaware Biotechnology Institute (DBI) suggested that the primers did not optimally amplify the target region. A TOPO® TA clone was performed on the product from the ethanol precipitation to amplify the target DNA for sequencing. This procedure uses Taq polymerase to clone DNA fragments into vectors using One Shot® Chemically Competent *E. coli* bacteria.

Sequencing was conducted at DBI's Sequencing and Genotyping Center with the state-of-the-art Applied Biosystems 3130 XL Genetic Analyzer, an automated capillary electrophoresis platform capable of analyzing 16 samples simultaneously. At the time of this writing, sequencing results from DBI are pending.

1.3.4 Phylogenetic analysis

This project undertook a morphological phylogenetic analysis using maximum parsimony. In accordance with the principle of parsimony, this method is based on the assumption that the best phylogenetic tree is the one that requires the least evolutionary change to explain relationships among taxa. It can be assumed that synapomorphies, or homology, occur more commonly than convergences, or homoplasy, so the most parsimonious phylogenetic tree is the one that minimizes homoplasy. Bootstrapping is an approach used to estimate support for clades on the tree; bootstrap values of 70% and higher are considered strongly indicative of true phylogeny. The program used for this component of the project was Phylogenetic Analysis Using Parsimony (PAUP* 4.0b10, Swofford 1998); tree graphics were developed using TreeView (Version 1.6.6, Page 1996).

For this phylogenetic analysis, both continuous and discrete characters were included. Although other studies have excluded traits that vary continuously for a variety of reasons, Poe and Wiens (2000) deem those reasons poorly founded and instead conclude that there is "nothing uniquely undesirable about continuous variation in phylogenetic analysis." Generally, Poe and Wiens (2000) advise that it is

preferable to err on the side of including possibly homoplastic character data than to discard potentially useful data. Following this guideline, characters with missing data were included; when data is missing for many taxa out of a small sample, this carries a risk of decreasing phylogenetic accuracy, but it is more likely to increase accuracy when data is only missing for a few taxa (Poe and Wiens 2000). Continuous characters (e.g., body length) were grouped in bins of roughly equal size (e.g., state 0 = 1.50-1.75 mm, state 1 = 1.76-2.00 mm, state 2 = 2.01-2.25 mm, and state 3 = 2.26-2.50 mm) and treated as ordered data, meaning that a difference between states 0 and 3 is more significant than a difference between states 0 and 1. For potentially overlapping traits (e.g., number of teeth on the calcar), the average number was used instead of the full range of possible numbers.

A total of 34 characters (Table 4), 14 ordered and 20 unordered, and 15 ingroup species were used in the MP analysis. Three dissimilar delphacid species, *Chionomus havanae, Kosswigianella lutulenta*, and *Muirodelphax arvensis*, were designated as the outgroup to root the tree. Successive weighting (Farris 1969) was performed on the MP tree until the topology stabilized. A bootstrap analysis (Felsenstein 1985) using importance sampling was performed to test phylogenetic support for tree topology.

The consistency index (CI), retention index (RI), homoplasy index (HI), rescaled consistency index (RC), and tree length were obtained for each tree. The CI, a measure of homoplasy, is calculated by adding the minimum number of steps across all characters, divided by the tree length, and tends to decrease as the number of

homoplasies rises. The RI, which measures the degree of potential synapomorphy on the tree in addition to homoplasy, is obtained by taking the product of the maximum number of changes on a tree and the number of changes on the tree divided by the product of the maximum number of changes on the tree and the minimum number of changes in the dataset. An RI of 0 represents the maximum amount of homoplasy, while an RI of 1 indicates no homoplasy. The RC is the product of multiplying the CI by the RI, and the HI is obtained by subtracting the CI from 1. The tree length is the minimum number of changes between character states needed to explain observed data in the tree.

1.3.5 Development of online resources

Representative specimens from included taxa were databased into the Plant Bug Planetary Biodiversity Inventory, a project developed by Toby Schuh of the American Museum of Natural History (<u>http://research.amnh.org/pbi/index.html</u>). A 2D-barcode label with a unique code, in the form of UDCC_NRI xxxxxxx, was added to each specimen. Databased specimens were representative of all localities for each species. Once included in the Plant Bug Inventory, the specimens were automatically added to the interactive maps on John Pickering's Discover Life website (<u>http://www.discoverlife.org/</u>).

Summarized morphological descriptions of the genus and each included species were developed for inclusion on the website for the overall project (<u>http://ag.udel.edu/enwc/research/delphacid/index.html</u>), along with notes on known

host plants, natural enemies, economic importance, photographs, molecular resources, biogeography and seasonality data, and selected references. This website is an ongoing effort to compile data about planthoppers for the purposes of crop protection against pest and invasive species, as well as broader academic interests.

1.4 Results

Out of 17 described species that were considered for inclusion in the newly revised *Caenodelphax*, 1 was retained in *Caenodelphax*, 8 were incorporated into a new genus, *Flavoclypeus*, 2 were treated as junior synonyms, and 6 were excluded from both genera. The species included in *Caenodelphax* and *Flavoclypeus* are redescribed in Chapter 2.

The unweighted heuristic search performed with PAUP* produced a single maximum parsimony tree (Figure 3). In this tree, the ingroup formed 3 clades: a basal clade of (aterrima + sucinea) + (recurvata + shermani) sister to the remaining ingroup; with the remaining ingroup forming two clades of (nitens + (balli + (teapae + livida)))) sister to the remaining 7 taxa. Successive weighting returned a tree with similar topology except for the movement of *Delphacodes nitens* to the base of the 7 taxon clade (Figure 4, Table 5). The bootstrap analysis yielded a finely resolved majority consensus tree (Figure 5).

Based on the tree generated through maximum parsimony analysis, eight of the species considered for inclusion in *Caenodelphax* form a monophyletic grouping with a bootstrap value of 66% on the basal node (Figure 5). *Caenodelphax teapae* did not

group with the remaining ingroup; consequently, monophyly of the expanded definition of *Caenodelphax sensu* Hamilton (2002) is not supported. *Caenodelphax* is here redefined as a monotypic genus, and the other 8 species forming a monophyletic grouping are here transferred to a new genus, *Flavoclypeus*.

Flavoclypeus nitens, new comb., is the basal taxon in the new genus and the least definitively placed. This species is placed outside the ingroup in the unweighted MP tree, but included in the ingroup in the weighted MP tree. Therefore, this species is tentatively included in *Flavoclypeus*.

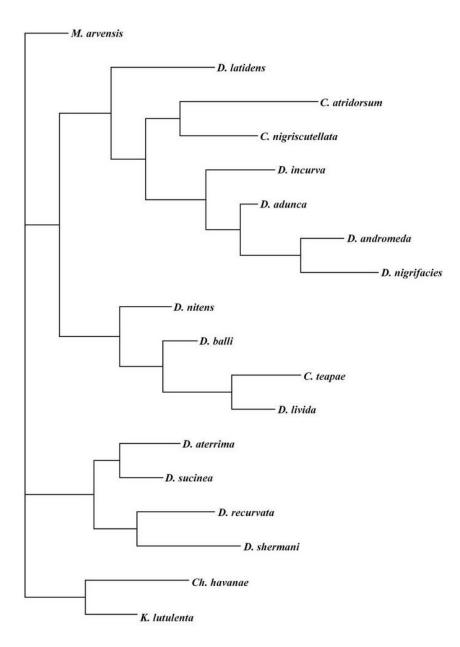


Figure 3. Unweighted MP tree of the *Caenodelphax* and *Delphacodes*-segregate ingroup and 3 outgroup species (*Chionomus havanae*, *Kosswigianella lutulenta*, and *Muirodelphax arvensis*). Tree length = 194, consistency index (CI) = 0.335, homoplasy index (HI) = 0.6649, retention index (RI) = 0.4241, rescaled consistency index (RC) = 0.1421.

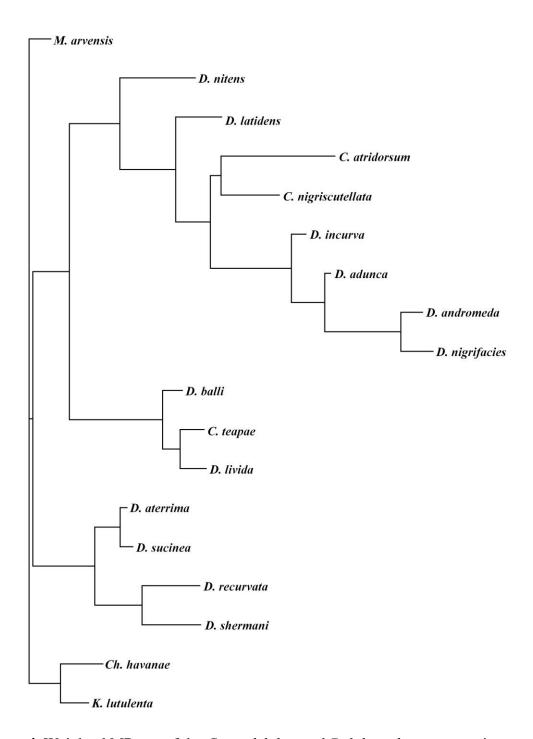
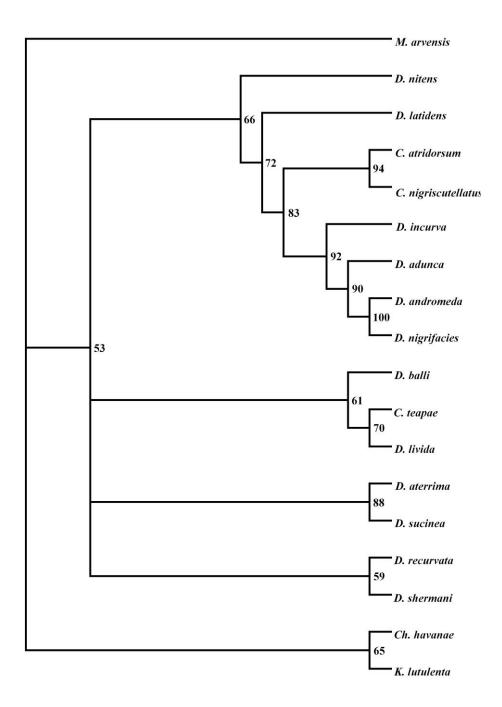


Figure 4. Weighted MP tree of the *Caenodelphax* and *Delphacodes*-segregate ingroup and 3 outgroup species (*Chionomus havanae*, *Kosswigianella lutulenta*, and *Muirodelphax arvensis*). Tree length = 23.86549, consistency index (CI) = 0.4615, homoplasy index (HI) = 0.5385, retention index (RI) = 0.6846, rescaled consistency index (RC) = 0.3159.



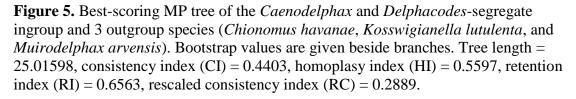


 Table 3. Morphological characters and states

Species	Character states	
Caenodelphax atridorsum	10011 10101 00101 31100 01011 01110 001	0
Caenodelphax nigriscutellatus	100?3 10111 00001 10210 11111 11110 001	0
Caenodelphax teapae	03110 12210 12112 01003 12110 01000 011	0
Chionomus havanae	011?0 10001 13201 00103 30110 000 000)1
Delphacodes adunca	12001 11211 01021 10310 02211 01011 101	0
Delphacodes andromeda	03001 01111 02030 30111 02211 01101 101	0
Delphacodes aterrima	13120 13211 02223 11200 ?001? 100 000	0
Delphacodes balli	033?2 12311 12211 11200 ??010 01110 001	0
Delphacodes incurva	22000 12211 01122 20210 ?120? 11110 001	1
Delphacodes latidens	11122 12111 01010 10212 01101 11100 001	0
Delphacodes livida	03110 12111 13011 11001 ?110? 001 001	0
Delphacodes nigrifacies	02001 01111 02001 00112 12001 001 001	0
Delphacodes nitens	12321 11111 02013 11201 11211 01110 011	0
Delphacodes recurvata	32331 11111 13023 11310 00100 001 000)1
Delphacodes shermani	32322 12210 03030 11312 ?201? 11010 011	0
Delphacodes sucinea	13121 13101 02021 21110 ?0000 000 000)1
Kosswigianella lutulenta	003?2 10001 11001 01201 0020? -00 000	0
Muirodelphax arvensis	20223 11211 12001 11112 ?0010 000 000	0

 Table 4. Character states for phylogenetic analysis

Table 4. Ch	laracter states for phylogenetic analysis
Body	
1.	Ratio of body length to width: 0) 2.40-2.65, 1) 2.66-2.90, 2) 2.91-3.15, 3) 3.16-3.40
2.	Ratio of body width to head width: 0) 2.60-3.00, 1) 3.01-3.40, 2) 3.41-3.80, 3) 3.81-4.25
3.	Length male brachypter: 0) 1.50-1.75mm, 1) 1.76-2.00 mm, 2) 2.01-2.25 mm, 3) 2.26-2.50 mm
4.	Length female brachypter: 0) 1.70-2.00 mm, 1) 2.01-2.30 mm, 2) 2.31-2.60 mm, 3) 2.61-2.90 mm
Head	
5.	Ratio of compound eye length to compound eye width: 0) 1.55-2.05, 1) 2.06-2.55, 2) 2.56-3.05, 3) 3.06-3.55
6. 7.	First and second antennal segments concolorous: 0) no, 1) yes Ratio of length of antennal segment I to width of antennal segment I: 0) 0.85-1.10, 1) 1.11-1.35, 2) 1.36-1.60, 3) 1.61-1.90

8.	Ratio of length antennal segment I to length antennal segment II: 0) 0.30- 0.40, 1) 0.41-0.50, 2) 0.51-0.60, 3) 0.61-0.70
9.	Carinae on frons concolorous: 0) no, 1) yes
9. 10.	Frons concolorous: 0) no, paler towards vertex, 1) yes
10.	Clypeus concolorous with frons: 0) no, 1) yes
11. 12.	Ratio of frons length to width: 0) 1.30-1.55, 1) 1.56-1.80, 2) 1.81-2.05, 3)
	2.06-2.35
13.	Vertex: 0) concolorous, 1) anterior compartments darker, posterior compartments lighter, 2) carinae paler
14.	Ratio of vertex length to width: 0) 0.85-1.05, 1) 1.06-1.25, 2) 1.26-1.45,
1	3) 1.46-1.65
15.	Ratio of length of anterior compartments of vertex to posterior
	compartments of vertex: 0) 0.60-0.80, 1) 0.81-1.00, 2) 1.01-1.20, 3) 1.21-
	1.45
16.	Ratio of vertex length to pronotum length: 0) 1.05-1.20, 1) 1.21-1.35, 2) 1.36-1.50, 3) 1.51-1.65
Thorax	
17.	Posterior edge of pronotum concolorous with mesonotum: 0) no, 1) yes
18.	Ratio of pronotum length to mesonotum length: 0) 0.35-0.45, 1) 0.46-
	0.55, 2) 0.56-0.65, 3) 0.66-0.75
19.	Wing color: 0) dark, 1) light
20.	Ratio of brachypter wing length to body length: 0) 0.35-0.45, 1) 0.46-
	0.55, 2) 0.56-0.65, 3) 0.66-0.75
21.	Average number of teeth on calcar: 0) 11.00-14.50, 1) 14.51-18.00, 2)
	18.01-21.50, 3) 21.51-25.00
Abdomen	
22.	Pygofer shape: 0) rectangular, taller than wide, 1) quadrate, roughly
	equally wide as tall, 2) triangular, taller than wide
23.	Dorsal emargination of the diaphragm shape: 0) concave, U-shape, 1)
	convex, ∩-shape, 2) W- or V-shape
24.	Median projection of the armature of the diaphragm: 0) absent, 1) present
25.	Armature of the diaphragm: 0) caudally projected, 1) dorsally projected
26.	Inner angles of parameres longer than outer angles: 0) no, 1) yes
27.	Processes on dorsal margin of segment 10: 0) absent, 1) present
28.	Processes on dorsal margin of segment 10: 0) short, 1) elongate
29.	Processes on dorsal margin of segment 10: 0) blunt or truncate, 1) sharp,
	pointed
30.	Processes on ventral margin of segment 10: 0) absent, 1) present
31.	Aedeagus: 0) projected caudally, 1) projected dorsally
32.	Aedeagus: 0) elongate, 1) stout
33.	Teeth on aedeagus: 0) absent, 1) present
34.	Processes on aedeagus: 0) absent, 1) present

Character	Туре	Weight	Character	Туре	Weight
1	Ordered	0.257143	18	Ordered	0.133333
2	Ordered	0.179487	19	Unordered	0.085714
3	Ordered	0.190476	20	Ordered	0.053254
4	Ordered	0.666667	21	Ordered	0.000000
5	Ordered	0.025000	22	Unordered	0.388889
6	Unordered	1.000000	23	Unordered	0.049383
7	Ordered	0.204545	24	Unordered	0.000000
8	Ordered	0.047619	25	Unordered	1.000000
9	Unordered	0.111111	26	Unordered	0.000000
10	Unordered	0.000000	27	Unordered	0.142857
11	Unordered	0.222222	28	Unordered	0.000000
12	Ordered	0.238095	29	Unordered	0.000000
13	Unordered	0.000000	30	Unordered	0.166667
14	Ordered	0.218750	31	Unordered	0.000000
15	Ordered	0.000000	32	Unordered	0.000000
16	Ordered	0.047619	33	Unordered	0.400000
17	Unordered	0.222222	34	Unordered	0.000000

Table 5. Character weights assigned by successive weighting by the rescaled consistency index in PAUP*.

Chapter 2

DESCRIPTIVE TAXONOMY

2.1 Caenodelphax Fennah, 1965

Type species: Liburnia teapae Fowler, 1905

Color. General body color glossy brown, patterned with orange or yellow; legs and antennae yellow; carinae concolorous or slightly darker. Genae often paler than frons; clypeus concolorous with frons. Anterior compartments of vertex usually darker than posterior compartments. Wings translucent, light to dark brown, often with darker venation. Females often paler.

Structure. Length 1.92-3.57 mm, with females larger. Carinae of head and thorax evident but concolorous with body. Antennae circular in cross-section, first segment longer than wide, second segment not quite twice as long as first. Head, including eyes, narrower than pronotum, vertex quadrate. Mesonotum more than twice as long as pronotum. Hind tibiae bearing 5 apical black teeth, grouped 2 + 3. Basitarsus with 7 apical black teeth grouped 2 + 5, and second tarsomere with 4 teeth. Calcar slender, acuminate, bearing continuous row of many fine, black-tipped teeth on outer margin. Wings in brachypters reaching nearly to end of abdomen. Genital diaphragm well-developed, armature projecting dorsocaudally, just broader than tall. Parameres broad, flattened. Segment 10 bearing 1 pair of processes.

Remarks. This genus bears a cursory resemblance to the new *Delphacodes*-segregate genus *Flavoclypeus* due to the pale antennae and contrasting darker frons, but can be distinguished by having the frons and clypeus concolorous instead of contrasting, genae contrasting with the frons instead of concolorous, and a higher frons length-to-width ratio.

Etymology. The genus name is presumably formed from the Greek adjective *caeno* meaning "sleek" or "shining" and the Greek noun *delphax* meaning "young pig". Fennah (1959) did not specify the etymological origin, but Fowler (1905) described the frons of *Liburnia teapae* as "more or less shining" in his original description. The name is masculine in gender based on the 1961 ICZN ruling (Opinion 602) that "*Delphax*" is masculine and consequently any name ending in "*-delphax*" is similarly considered masculine.

2.1.1 Caenodelphax teapae (Fowler, 1905)

Liburnia teapae Fowler, 1905: 135.

Megamelus teapae (Fowler); combination by Crawford, 1914: 618.

Delphacodes teapae (Fowler); combination by Wolcott, 1923: 274.

Delphacodes philyra Fennah, 1959: 262.

Caenodelphax teapae (Fowler); combination by Fennah, 1965: 96. *Caenodelphax philyra* (Fennah); combination by Fennah, 1965: 96.

Caenodelphax philyra (Fennah); new synonymy

Type locality. Mexico, Tabasco state, Teapa.

Diagnosis. General body color glossy brown with yellow to orange antennae, patches on genae, and legs; wings translucent dark brown. Length 1.92-3.57 mm, varied by sex and wing morph. Parameres broad, constricted most narrowly subapically, truncate to slightly concave apically. Aedeagus tapering from broad base to rounded apex, bearing irregular row of about 5 retrose teeth on apical half. Segment 10 bearing pair of short, blunt, ventrocaudally curved processes.

Color. General body color glossy brown to dark brown, carinae concolorous with body. Genae paler; antennae and legs yellow, darker near articulation of femur and coxa. Wings translucent dark brown except clear at apex of clavus and cubital veins; veins dark. Pygofer brown. Females may display similar coloration to males but typically appear paler (see remarks).

Structure. Length ♂ macropter: 2.62 mm (2.05-3.15, n= 10); length ♀ macropter:
3.11 mm (2.90-3.57, n = 5). Length ♂ brachypter: 1.92 mm (n = 1); ♀ brachypter:
2.07 mm (2.04-2.10; n = 2).

Head. Head, including eyes, slightly narrower than prothorax. Frons quadrate, roughly twice as long as wide (1:w 2.04:1); strong median and lateral carinae; lateral carinae subparallel. Median carina of frons forked below fastigium. Vertex approximately as wide as long (1:w 1.11:1), carinae evident. Antennal segment I longer than wide (1:w 1.57:1); second antennal segment approximately twice as long as first (I:II 0.58:1), bearing sensory fields arranged approximately in rows.

Thorax. Mesonotum more than twice as long as pronotum (pronotum l:mesonotum l 0.38:1); pronotum and mesonotum weakly carinate. Lateral carinae of pronotum curved lateral, not reaching posterior margin. Median carina of mesonotum becoming obsolete in scutellum, lateral carinae slightly diverging posteriorly, reaching hind margin. Wings rounded at apex, veins setose. Calcar flattened, widest in basal third, slightly narrowing distally to acute apex, roughly three-quarters length of basitarsus, bearing a continuous row of 13-18 (n = 6) fine, black-tipped teeth on outer margin.

Abdomen. Pygofer approximately triangular in lateral view, much wider ventrally than dorsally; in caudal view, opening taller than broad. Parameres wide, approximately parallel, broad basally, basal angles evident, not projected; distally narrowed then becoming broader at truncate to slightly concave apex; inner angles elongate, pointed medially. Suspensorium ring-shaped. Aedeagus tubular, somewhat flattened, in lateral view broadest proximally, narrowed distally to rounded apex; bearing an irregular row of about 5 small lateral teeth on left side arranged diagonally from subapical dorsal

margin to ventral margin near half length of aedeagus. Segment 10 in lateral view taller than long, bearing pair of broad, short, blunt, curved processes on ventrolateral margin; processes serrulate apically. Segment 11 elongate, nearly as long as height of segment 10.

Hosts.

Axonopus compressus (Sw.) P. Beauv. (broadleaf carpetgrass) (Fennah 1959)

Crotalaria L. (rattlebox) (Leonard 1933)

Cucurbita maxima Duchesne (winter squash) (NMNH, Puerto Rico)

Cymbopogon citratus (D.C. ex Nees) Stapf (lemon grass) (Wolcott 1923)

Cynodon dactylon (L.) Pers. (Bermudagrass) (NMNH, Puerto Rico)

Daucus L. (carrot) (Wolcott 1923)

Paspalum notatum Flueggé (bahiagrass) (NMNH, Florida)

Phaseolus vulgaris L. (kidney bean) (NMNH, Puerto Rico)

Saccharum L. (sugarcane) (Wolcott 1923)

Solenostemon scutellarioides (L.) Codd (common coleus, Lamiaceae; reported as Coleus blumei) (Ballou 1936)

Urochloa plantaginea (Link) R. Webster (plantain signalgrass) (Wilson 2005)

Distribution. USA (FL); Caribbean (Antigua, Cuba, Dominica, Dominican Republic, Grenada, Haiti, Jamaica, Martinique, Puerto Rico, Saint Lucia, Trinidad and Tobago); Bahamas, Belize, Bolivia, Brazil, Colombia, Costa Rica, Ecuador, El Salvador, French Guiana, Guadeloupe, Guatemala, Guyana, Honduras, Mexico (Jalisco, Tabasco,
Veracruz), Nicaragua, Panama, Peru, Venezuela; also reported from Barbados (Fennah 1965), Mexico (Guerrero, Puebla, San Luis Potosí) (PBI database; UKYC),
Montserrat (Fennah 1959), St. Thomas and St. Croix (Caldwell and Martorell 1951),
St. Vincent and the Grenadines (Fennah 1959).

Remarks. This is a broadly distributed and very common Neotropical species, often collected while sweeping grasses or at lights. Most individuals are macropterous, although brachypters are not uncommon, particularly among females. There is variation in coloration geographically as well as sexually; females are usually paler, with general body color yellowish to light brown, having a yellow vertex and frons gradually darkening to brown towards the clypeus; carinae yellow, intercarinal regions brown. The wings of females usually have more extensive clear regions. Brachypters are usually paler than macropters. Crawford (1914) observed that specimens from Cuba were more uniformly brown than black. He designated the variety *Megamelus teapae albinotatus* from specimens collected in Jalapa, Mexico, but this variety was subsequently raised to the species level (*Delphacodes albinotata*) by Muir and Giffard (1924) and later determined to be *Peregrinus maidis* (Ashmead) by Beamer (1948b). Wolcott (1950) observed that *C. teapae* is frequently preyed upon by the lizards *Anolis pulchellus* and *A. krugii*.

Caenodelphax teapae can be distinguished easily from the closely-allied species in the genus *Flavoclypeus* by the clypeus, which is concolorous with the frons,

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not paler. The genae are often paler than the frons, in contrast to members of *Flavoclypeus*, which have the genae and frons concolorous. Compared to the sympatric tropical taxa in *Flavoclypeus* (*F. andromedus* and *F. nigrifacies*), *C. teapae* has darker wings and a darker posterior edge of the pronotum.

The lectotype is a macropterous male in good condition, but it is glued on a card that obscures some features. Fennah (1967) designated the lectotype from the original series described by Fowler in 1905.

Fennah's 1959 description of *Delphacodes philyra* outlined structural differences between it and *D. teapae*, but these differences fall within the realm of normal geographic variation within *C. teapae*. Fennah mentioned the calcar of *D. philyra* bearing 18 teeth in contrast to *D. teapae*'s 13; this analysis found the number of calcar teeth in *C. teapae* to be more variable (13-18), and Fennah (1965) reported up to 21 teeth. Fennah (1965) cited differences in coloration of the vertex as a key component of the distinction between the two species; further review indicates that *C. teapae* displays high variation in coloration in many parts of the body. Additional reported differences, involving the processes of segment 10 and the arrangement of spines on the aedeagus, were deemed to be less pronounced than Fennah originally described them, and well within the scope of intraspecific geographic variation. The fact that *C. philyra* is only known from one collecting event, despite extensive Neotropical sampling efforts, further lends support to its being a junior synonym of *C. teapae*.

Etymology. The specific name is a reference to the locality of the type specimen, (Teapa, Mexico).

Type material examined. MEXICO: Tabasco: Lectotype (male macropter, BMNH): "Type / H. T. [round label, red border] // Lecto- / type [round label, blue border] // Liburnia / teapae / Fowler [handwritten, paper folded] // Teapa, / Tabasco. / H. H. S[mith]. // B.C.A. Homopt. I / Liburnia / teapæ, / Fowl." ST. LUCIA: Holotype *D*. *philyra* (male macropter, BMNH): "Type [round label] // Pres by / Com Inst Ent / B M 1965-3 // Morne Fortunée [handwritten] / St. Lucia W. I. / Feb. 1940 / R. G. Fennah // Delphacodes / philyra Fenn. / det / RGFennah / TYPE".

Other material examined. See Appendix A.

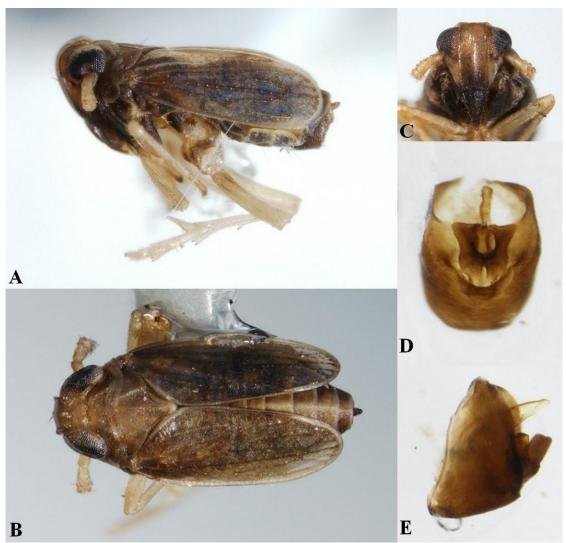


Figure 6. Features of *Caenodelphax teapae*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2 Flavoclypeus, New Genus

Type species: Liburnia andromeda Van Duzee, 1907

Color. General body color glossy dark brown to black, often with white to yellow patterning, carinae concolorous; antennae, clypeus, and legs white to yellow. Genae concolorous with frons. Vertex concolorous except in *F. incurvus*, which has the anterior compartments darker than the posterior compartments. Wings usually clear, light; dark in *F. atridorsum* and *F. nitens*. Females generally paler, often uniformly white, yellow, or tan.

Structure. Length 1.41-3.64 mm, with females larger. Carinae evident, concolorous with body. Antennae circular in cross-section, first segment approximately as wide as long, half length of second segment. Head, including eyes, narrower than pronotum. Mesonotum not quite twice as long as pronotum. Hind tibiae bearing 5 apical black teeth, grouped 2 + 3. Basitarsus with 7 apical black teeth grouped 2 + 5, and second tarsomere with 4 teeth. Calcar slender, acuminate, bearing continuous row of fine, black-tipped teeth on outer margin. Wings in brachypters leaving several abdominal tergites exposed. Diaphragm well-developed, often with median projection, dorsally or dorsocaudally projecting. Segment 10 bearing one or two pairs of processes on posterior margin.

Remarks. This is a broadly distributed New World genus with some common, easily encountered species and some rarer, more cryptic species. Several species in this genus are externally nearly identical, necessitating examination of the male genitalia for clear species determination. Although the yellow clypeus and dark frons pattern is a useful identifying feature of this genus, it is not sufficiently diagnostic in itself as several *Delphacodes* species (*D. aterrima*, *D. balli*, and *D. shermani*) share this feature.

Etymology. The generic name is formed from the Latin adjective *flavo* meaning "yellow" and the Latin noun *clypeus* meaning "shield" in reference to the pale clypeus, which is in sharp contrast to the dark frons observed in the included species. The name is treated as masculine in gender.

2.2.1 Key to the males of *Flavoclypeus*

	One pair of processes on segment 10
4.	First pair of processes on segment 10 short and slender; first antennal segment
	yellowF. aduncus
	First pair of processes on segment 10 elongate, broad, spatulate; first antennal
	segment brown F. andromedus
5.	Inner angles of parameres more pronounced than outer angles; pronotum
	mostly pale; first antennal segment yellow6
	Outer angles of parameres slightly longer than inner angles; pronotum mostly
	dark except for posterior edge; first antennal segment brown F. nigrifacies
6.	Inner angles of parameres strongly evident, but not elongate7
	Inner angles of parameres elongate F. nigriscutellatus
7.	Processes of segment 10 truncate apically, ventrocaudally projecting; vertex
	concolorous, or with pale posterior edgeF. latidens
	Processes of segment 10 sharply incurved apically, terminating in pointed
	apices; posterior compartments of vertex paler than anterior compartments
	F. incurvus

2.2.2 Flavoclypeus andromedus (Van Duzee, 1907), new comb.

Liburnia andromeda Van Duzee, 1907: 46.

Megamelus andromedus (Van Duzee, 1907); combination by Crawford, 1914: 628.

Delphacodes andromeda (Van Duzee, 1907); combination by Muir and Giffard, 1924: 36

Type locality. Jamaica, Middlesex County: Mandeville.

Diagnosis. General body color glossy dark brown, with yellow to orange clypeus, second antennal segment, legs, and posterior edge of pronotum; wings clear. Length 1.45-2.79 mm, varied by sex and wing morph. Armature of the diaphragm W-shaped. Parameres flattened, broadest apically. Aedeagus tubular, broadest subapically, bearing 6 teeth near gonopore. Segment 10 bearing 2 pairs of processes; first pair broad, spatulate; second pair elongate, sinuous.

Color. Frons glossy dark brown to black with sharply contrasting yellow to orange clypeus; first antennal segment brown, second antennal segment yellow to orange. Thorax dark brown to black, carinae darkest. Legs pale yellow to orange-brown, palest distally, darker towards coxae. Posterior edge of pronotum and posterior tip of mesonotum pale white to yellow. Abdomen fading from dark brown anteriorly to pale white or yellow posteriorly; posterior edge of each abdominal segment slightly darker than anterior edge. Wings translucent, veins indistinct. Pygofer dark brown to black, with paler spot on dorsum. Females may be similar in coloration to males or may appear paler, uniform yellow.

Structure. Length \bigcirc macropter: 2.64 mm (2.47-2.79, n = 3); length \bigcirc macropter: 2.57 mm (n = 1). Length \bigcirc brachypter: 1.52 mm (1.45-1.57, n = 8); length \bigcirc brachypter: 1.83 mm (1.67-1.94, n = 13).

Head. Head, including eyes, slightly narrower than prothorax. Frons approximately twice as long as broad (l:w 1.81:1), widest at middle; carinae strongly evident. Vertex longer than wide (l:w 1.56:1). Antennal segment I approximately equal in length and width (l:w 1.13:1); second antennal segment approximately twice as long as first (I:II 0.47:1).

Thorax. Mesonotum about twice as long as pronotum (pronotum 1:mesonotum 1 0.55:1); mesonotum and pronotum strongly carinate. Median carina of mesonotum becoming obsolete on scutellum. Wings rounded at apex, about twice as long as wide in brachypters, extending for one-third length beyond abdomen in macropters. Abdominal segments 7-10 visible dorsally in brachypters. Calcar approximately three-quarters length of basitarsus, foliaceous, bearing row of 12-15 (n = 5) fine, black-tipped teeth on outer margin.

Abdomen. Pygofer roughly triangular in lateral view, much wider ventrally than dorsally; anterior margin longer than posterior margin; in caudal view, aperture approximately oval. Diaphragm well-developed; armature of the diaphragm projecting dorsally, approximately W-shaped with rounded medial projection, taller than wide. Parameres broad basally, narrowing slightly medially, and broadest apically. Aedeagus tubular, in lateral view broadest subapically, with row of approximately 6 teeth surrounding gonopore at apex. Segment 10 bearing 2 pairs of processes; first pair distinctly flattened, spatulate, broadest basally; second pair elongate, sinuous, arising on caudal margin of segment 10, projecting dorsally.

Hosts.

Paspalum L. (crowngrass) (Osborn 1926)Eleocharis R. Br (spikerush; see remarks) (UDCC, Delaware)

Distribution. USA (AL, AR, DE, FL, LA, MD, NC, NJ, PA, TN, TX, VA), Jamaica. Also reported from USA (CT) (PBI database; NCSU), USA (GA) (Spooner 1920), USA (KS) (PBI database; NCSU), USA (KY) (PBI database; NCSU), USA (MA) (PBI database; NCSU), USA (OH) (Osborn 1935), USA (OK) (PBI database; OSEC), USA (SC) (PBI database; NCSU), Belize (Crawford 1914), Cuba (Osborn 1926), Guyana (Van Duzee 1907), Puerto Rico (Osborn 1929).

Etymology. The specific name is presumably related to the Latin *Andromeda* (Latinized form of the Greek *Andromede*), beautiful Ethiopian princess in the popular Greek myth. Van Duzee (1909) referred to *L. andromeda* as a "beautiful little species".

Remarks. Osborn (1935) comments that this species occurs in high numbers in moist locations. According to label data, an additional host for this species might be *Eleocharis* R. Br (spikerush) but this remains unconfirmed (see Appendix A).

Although the type specimen of this species was collected in Jamaica, this species does not have a broad Neotropical distribution. Specimens collected in South America should be checked against *F. nigrifacies* as these species can easily be mistaken for one another, although the latter has only one pair of processes on segment 10. As noted above, this species also bears a resemblance to *F. aduncus*, but with the first pair of processes on segment 10 longer and spatulate rather than short and sharply pointed.

Type material examined. JAMAICA: "Mandev'le / Ja. Apr. 06 // VanDuzee /Collector // [handwritten] // [red paper] LECTOTYPE andromeda // EPVanDuzee /Collection // California Academy / of Sciences / Type No. 3059".

Material examined. See Appendix A.

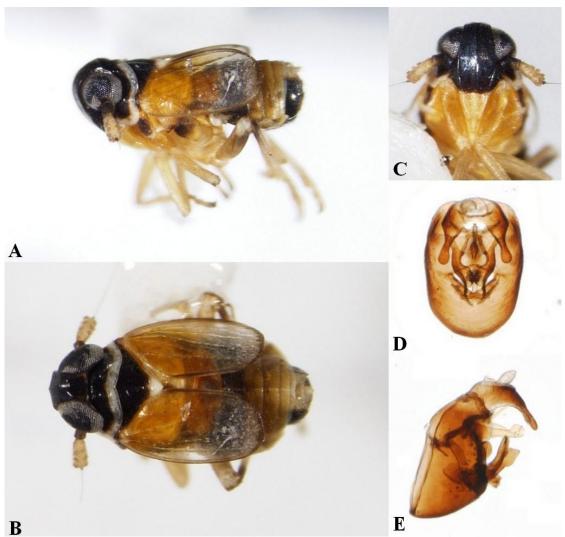


Figure 7. Features of *Flavoclypeus andromedus*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.3 Flavoclypeus aduncus (Beamer 1948), new comb.

Delphacodes adunca Beamer 1948b: 98.

Type locality. US, Florida: Levi County.

Diagnosis. General body color glossy dark brown with white to yellow antennae, clypeus, legs, and posterior edge of pronotum; wings clear. Length 1.41-1.91 mm. Parameres broad, boot-shaped. Aedeagus tubular, bearing many retrose spines, bent dorsally near base, bent caudally in apical third. Segment 10 bearing 2 pairs of processes; first pair short, sharply pointed; second pair elongate, sinuous.

Color. Frons dark brown to black with sharply contrasting yellow antennae and clypeus. Thorax dark brown to black, legs white to yellow. Posterior edge of pronotum and scutellum usually pale white to yellow. Abdomen fading from dark brown anteriorly to pale white or yellow posteriorly. Wings translucent, veins indistinct. Pygofer, in dorsal view, white to yellow medially and dark brown to black laterally. Sexually dimorphic coloration with females typically paler, uniform white to yellow.

Structure. Macropters: none observed. Length δ brachypter: 1.61 mm (1.41-1.91, n = 20); \mathcal{Q} brachypter: none observed.

Head. Head, including eyes, slightly narrower than prothorax. Frons approximately twice as long as wide (l:w 1.80:1). Vertex longer than wide (l:w 1.38:1), carinae evident. Antennal segment I slightly longer than wide (l:w 1.13:1); second antennal segment approximately twice as long as first (I:II 0.53:1), bearing sensory fields arranged approximately in rows.

45

Thorax. Mesonotum longer than pronotum (pronotum 1:mesonotum 1 0.69:1); pronotal and mesonotal carinae evident. Median carina of mesonotum becoming obsolete in scutellum, lateral carinae slightly diverging posteriorly, reaching hind margin. Wings rounded at apex. Calcar flattened, acuminate, widest in basal third, approximately three-quarters length of basitarsus, slightly narrowing distally to acute apex, bearing a continuous row of 10-13 (n = 5) fine, black-tipped teeth on outer margin.

Abdomen. Pygofer approximately ovular in lateral view, slightly wider ventrally than dorsally; anterior margin longer than posterior margin. Diaphragm strongly developed, in caudal view armature deeply concave, just taller than wide, dorsocaudally projected; not visible in lateral view. Parameres broad, in caudal view boot-shaped, inner angles strongly evident. Segment 10 with two pairs of processes; the first short, caudally projected, terminating in sharp apices; the second more elongate, sinuous, and dorsally projected, terminating in sharp apices. Aedeagus tubular, bearing numerous retrose spines; bent dorsally in basal third; bent in apical third to project caudally; slightly wider at base than at apex, narrowest medially.

Hosts. None reported.

Distribution. USA (FL, GA, NC).

Etymology. Beamer (1948b) did not specify the etymological origin, but the specific name is presumably formed from the Latin adjective *aduncus* meaning "hooked" or "bent inward", in reference to the aedeagus, which is bent dorsally near the base.

Remarks. This species is very similar to *F. andromedus* in coloration and structure. Both species bear 2 processes on segment 10, but in *F. aduncus*, the first pair is short and sharply pointed, whereas it is elongate and spatulate in *F. andromedus*.

Type material examined. USA: FLORIDA: Paratypes (2 male brachypters, USNM), "Hilliard FLA / Oct5 / Oman 1938 // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer", paratype (female brachypter, USNM): "Islamoranda / FLA July 20 / Oman 1939 // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer", (2 male brachypters, USNM): "LaBelle FLA / July 16 / Oman 1939 // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer", (2 male brachypters, USNM): "New Port Ritchey / FLA X-7 / Oman // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer", (male brachypter, USNM): "Sanford, Fla. / (handwritten)10-31-25 / E. D. Ball // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer", (2 male brachypters, USNM): "ZolfoSpgs / FLA Ju. 15 / Oman 1939 // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer", USA: NORTH CAROLINA: (3 male brachypters, USNM), "Raleigh NC / Oct 16 / Oman 1938 // [blue paper] PARATYPE / Delphacodes / adunca / R.H. Beamer". Other material examined. USA: GEORGIA: <u>Rabun Co</u>., Pinnacle Mt., 2500-3000', 20 August 1913 (1 broken, USNM), <u>Thomas Co</u>., Thomasville, 21 April 1914 (1♀, USNM), 9 April 1915, C.S. Spooner (4♂, USNM), 10 April 1915 (1♂, USNM), 11 April 1915, C.S. Spooner (2♂, USNM), 15 April 1915 (1♂, USNM), 22 April 1915 (1♂, USNM), 4 May 1915 (1♂, USNM). NORTH CAROLINA: <u>Wake Co</u>., Raleigh, 19 June 1993, C.R. Bartlett (1♂, UDCC).

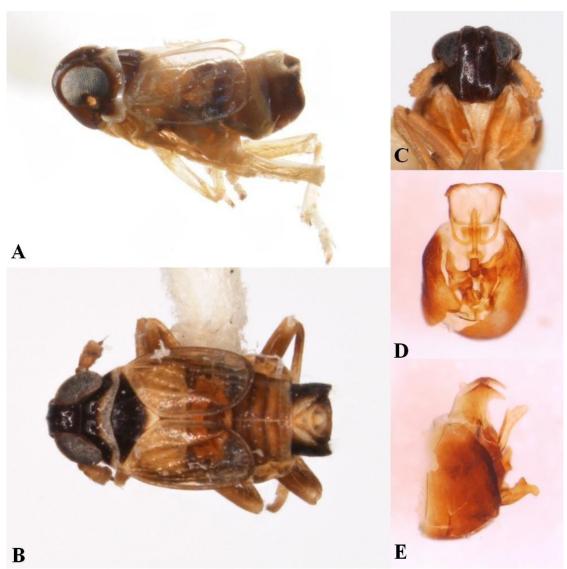


Figure 8. Features of *Flavoclypeus aduncus*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.4 Flavoclypeus atridorsum (Beamer 1947), new comb.

Delphacodes atridorsum Beamer 1947: 63.

Caenodelphax atridorsum (Beamer 1947), combination by Hamilton (2002:

17)

Type locality. US, Oregon: Deschutes County.

Diagnosis. General body color dark brown to black with yellow clypeus, antennae, and legs; wings dark. Length 1.66-2.23 mm, with females larger. Parameres broad, with inner angles sharply pointed medially. Aedeagus bent dorsally at base, bearing about 6 teeth near apex and about 8 teeth near base. Segment 10 bearing pair of pointed processes curved slightly ventrally.

Color. General body color dark brown to black (males), carinae concolorous with body. First and second antennal segments, clypeus, legs, and scutellum yellow; genae fading from dark brown to yellow. Wings dark, translucent brown, veins darker. Pygofer dark brown to black. Sexually dimorphic coloration with females paler, general body color uniform yellow to light brown, with lateral frontal carinae and apex of ovipositor darker; in some specimens, posterior edge of each abdominal segment darker than anterior edge.

Structure. Macropters: none observed. Length 3° brachypter: 1.69 mm (1.66-1.71, n = 2); length 2° brachypter: 2.04 mm (1.89-2.23, n = 5).

Head. Frons slightly longer than wide (l:w 1.33:1). Frons widest at middle, between ocelli, and tapering evenly towards the vertex above and clypeus below. Median carina defined most sharply medially, fading at base and apex. Antennal segment I

approximately equal in length and width (1:w 0.88:1); second antennal segment approximately twice as long as first (I:II 0.44:1). Vertex as long as wide (1:w 1:1), broadly rounded in frontal view.

Thorax. Mesonotum approximately twice as long as pronotum (pronotum 10.53:1); pronotal and mesonotal carinae visible but not strongly evident. Median carina of mesonotum becoming obsolete before scutellum; lateral carinae diverging sharply towards hind margin. Wings rounded, only slightly longer than wide, just reaching second abdominal segment. Calcar foliaceous, approximately three-quarters length of basitarsus, bearing a row of 10-12 (n = 3) very fine, black-tipped teeth on outer margin.

Abdomen. Pygofer approximately quadrate in lateral view, slightly taller than wide. Segment 10 bearing a pair of sharp, pointed processes curved slightly ventrally. Parameres broad basally and apically, tapering to narrowest point medially; inner angles strongly evident, pointing medially. Suspensorium ring-shaped. Aedeagus strongly projected dorsally, broadest basally but tapering only slightly towards apex, with about 6 teeth located apically and about 8 located basally.

Hosts. None reported.

Distribution. USA (OR).

51

Etymology. Beamer (1947) did not specify the etymological origin of the specific name, but it was likely formed from the Latin *ater* meaning "black" and the Latin noun *dorsum* meaning "back" in reference to the dark body color observed in males.

Remarks. As Beamer (1947) noted, this species is similar to *F. nitens* but smaller and bearing a distinctive bend in the aedeagus. In frontal view, *F. atridorsum*'s head is rounder than those of the other species in *Flavoclypeus* due in part to the shorter length of the frons.

This species is only known to have been collected during the month of July.

Material examined. USA: OREGON: <u>Deschutes Co.</u>, Lapine, 2 July 1935, Oman $(2^{\circ}, 5^{\circ}, \text{USNM})$.

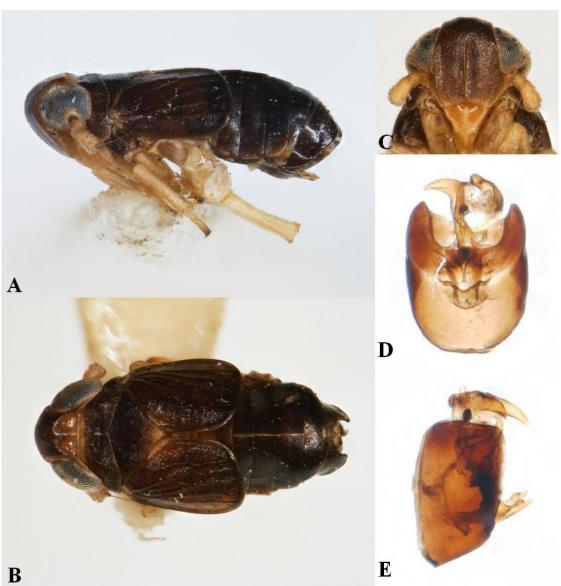


Figure 9. Features of *Flavoclypeus atridorsum*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.5 Flavoclypeus incurvus (Beamer 1948), new comb.

Delphacodes incurva Beamer 1948a: 3.

Type locality. US, Connecticut: Tolland County.

Diagnosis. General body color glossy dark brown; clypeus, antennae, posterior edge of pronotum, and legs white to yellow; wings clear. Length 1.45-1.96 mm, with females larger. Parameres avicephaliform, with sharp inner angles and rounded outer angles. Aedeagus elongate, bearing small teeth near apex and 3 larger processes medially on ventral margin. Segment 10 bearing pair of processes on dorsal margin with apices sharply incurved medially.

Color. General body color glossy dark brown, carinae concolorous with body. First and second antennal segments, clypeus, posterior edge of vertex, pronotum, scutellum, and legs white to yellow. Wings translucent, veins whitish. Pygofer brown, with paler medial spot in dorsal view. Females paler; general body color uniformly white to yellow.

Structure. Length \bigcirc brachypter: 1.57 mm (1.45-1.67 mm, *n* = 5); \bigcirc brachypter: 1.83 mm (1.52-1.96 mm, *n* = 5).

Head. Width of head, including eyes, subequal to prothorax width. Frons longer than wide (1:w 1.59:1); facial carinae evident. Vertex longer than wide (1:w 1.38:1), broadly rounded in frontal view. Antennal segment I longer than wide (1:w 1.5:1); second antennal segment approximately twice as long as first (I:II 0.53:1).

Thorax. Mesonotum not quite twice as long as pronotum (pronotum 1:mesonotum 1 0.59:1), carinae evident. Median carina of mesonotum becoming obsolete in scutellum. Wings rounded at apex. Calcar knife-shaped, bearing continuous row of 6-13 (n = 5) very fine, black-tipped teeth.

Abdomen. Pygofer approximately quadrate in lateral view, width and height roughly equal, anterior margin just taller than posterior margin. Parameres avicephaliform, broadest apically, narrowest just before apex, with inner angles strongly produced into sharp apices, outer angles rounded. Aedeagus tubular, broadest basally, narrowest medially, incurved ventrally at apex, bearing irregular row of teeth apically and 3 larger, ventrally-projected processes at midlength of ventral margin. Segment 10 with pair of elongate, caudally-projected processes on dorsal margin, terminating in pointed apices incurved medially at right angles.

Hosts. None reported.

Distribution. USA (CT, NM, UT). Also reported from USA (KS) (Beamer 1948a), Canada (BC) (Maw et al. 2000).

Etymology. Beamer (1948a) did not specify the origin of the specific name, but it is likely in reference to the sharply incurved processes of segment 10.

Remarks. As Beamer (1948a) noted, this species is very similar to *F. nigriscutellatus* but with the posterior compartments of the vertex paler than the anterior compartments; this feature also helps to distinguish it from *F. aduncus*. In contrast to both of those species, *F. incurvus* has the anterior compartments of the vertex longer than the posterior compartments, as well as the dorsal processes on segment 10 sharply incurving medially.

Type material examined. **USA**: CONNECTICUT: Paratype (female brachypter, AMNH): "Storrs Conn. / 8-15-1946 / R. H. Beamer // [blue paper] PARATYPE / Delphacodes / incurva / R. H. Beamer".

Other material examined. USA: NEW MEXICO: Marshall, 26 July 1950, D.D. Beamer (1 \bigcirc , SEMC), <u>Colfax Co.</u>, Maxwell, 26 July 1950, R.H. Beamer (3 \bigcirc , 7 \bigcirc SEMC). UTAH: <u>Uintah Co.</u>, Vernal, 2 August 1947, R.H. Beamer (7 \bigcirc , 1 \bigcirc , SEMC).

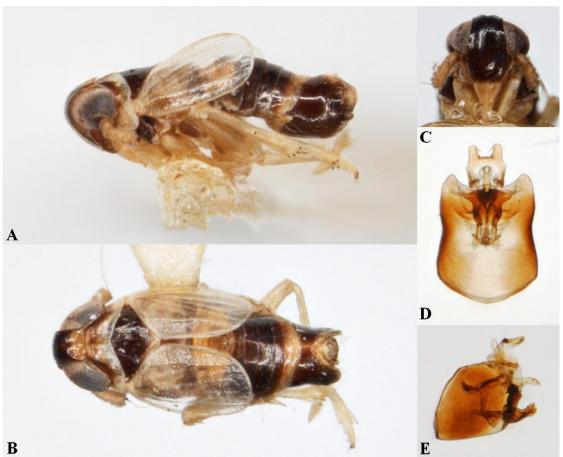


Figure 10. Features of *Flavoclypeus incurvus*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.6 Flavoclypeus latidens (Beamer 1948), new comb.

Delphacodes latidens Beamer 1948a: 4.

Type locality. US, Texas: Kenedy County.

Diagnosis. General body color glossy dark brown, with white to yellow clypeus,

antennae, pronotum, and legs; wings clear. Length 1.81-3.64 mm, varied by sex and

wing morph. Parameres broad, avicephaliform, with elongate inner angles pointed medially. Aedeagus tapering from broad base to narrow apex, with two rows of retrose teeth. Segment 10 bearing pair of thick, truncate processes.

Color. General body color glossy dark brown, carinae concolorous with body. First and second antennal segments, clypeus, lower margin of frons, posterior edge of vertex, pronotum, scutellum, and legs white to yellow. Wings translucent, veins light brown. Pygofer brown, with paler medial spot in dorsal view. Females paler; general body color yellow, with pale median vitta and row of dark brown to black patches occurring along the dorso-lateral margins of the abdomen and dark patches on genae, directly below the compound eyes.

Structure. Length 3 macropter: 2.97 mm (2.86-3.36, n = 5); 2 macropter: 3.40 mm (3.21-3.64, n = 5). Length 3 brachypter: 1.89 mm (1.88-1.98, n = 6); length 2 brachypter: 2.49 mm (2.38-2.61, n = 4).

Head. Head, including compound eyes, slightly narrower than prothorax. Frons not quite twice as long as broad (l:w 1.70:1); facial carinae strongly evident, median carina of frons forked below fastigium. Vertex approximately as long as wide in males (l:w 1.11:1); slightly longer in females. Antennal segment I longer than wide (l:w 1.38:1); second antennal segment twice as long as first (I:II 0.50:1).

Thorax. Mesonotum approximately twice as long as pronotum (pronotum l:mesonotum l 0.56:1), carinae evident. Median carina of mesonotum becoming obsolete in scutellum; lateral carinae diverging posteriorly, just reaching hind margins. Wings rounded at apex. Calcar bearing continuous row of 12-19 (n = 5) teeth.

Abdomen. Pygofer approximately quadrate in lateral view, slightly wider ventrally than dorsally. Diaphragm strongly developed; armature with rounded dorsocaudal projection, just wider than tall. Parameres wide basally and apically, narrowest medially, approximately avicephaliform; inner angles elongate, projected medially, outer angles acute, projected laterally. Aedeagus widest basally, tapering slightly to thinnest point apically; retrose teeth occurring approximately in one row of 2-4 dorsally, closer to apex than base, and one row of 6-8 ventrally, closer to base than apex. Segment 10 bearing one pair of thick, truncate processes, ventrocaudally projected, apices slightly laterally projected. Segment 11 approximately ovular, longer than wide.

Hosts.

Setaria texana W.H.P. Emery (Texas bristlegrass) (Wilson et al. 1994)

Distribution. USA (AZ, KS, NM, TX, UT), Mexico. Also reported from USA (OK) (Wilson and Wheeler 2010).

Etymology. Beamer (1948a) did not specify the origin of the specific name, but it was presumably formed from the Latin adjective *latus* meaning "broad" and the Latin noun *dens* meaning "tooth" in reference to the aedeagal teeth.

Remarks.

This species bears a superficial resemblance to *F. nigriscutellatus* in coloration, as both species' pronota are pale, but can be distinguished by the genitalia. *F. nigriscutellatus* has more elongate inner angles on the parameres and a curved, slightly pointed aedeagus, compared to avicephaliform parameres and a blunt, caudally-projected aedeagus in *F. latidens*.

This species has been collected from April through September. The record from Kansas is tentative because it is based on a female specimen only.

Material examined. USA: ARIZONA: <u>Cochise Co.</u>, Chiricahua Mountains, Rucker Camp, T. 29S. R. 29E. Sec. 27, 4-7 September 1987, pan trap, T.D. Miller (1♂, UDCC), Huachuca Mountains, Garden Canyon Upper Picnic Area, 7 May 2009, swept seep area, C.W. and L.B. O'Brien (1 broken, UDCC), <u>Pima Co.</u>, Baboquivari Mountains, 11 April 1932, E.D. Ball (6♂, 4♀, USNM), Green Valley, 3107', N31.80, W111.03, UV light, 25 August 2007, J. Brambila (1♂, UDCC), <u>Santa Cruz Co.</u>, Nogales, Peña Blanca Lake, 12 September 2008, C.W. O'Brien (6♂, 31♀, UDCC), Nogales, Peña Blanca Lake, Boat Ramp Area, 5 June 2005, L.B. and C.W. O'Brien (1♂, UDCC), Nogales, Peña Blanca Lake, Upper White Rock Campground, 12 September 2008, C.W. O'Brien ($3^{\circ}_{\circ}, 9^{\circ}_{\circ}$, UDCC). KANSAS: Meade Co., junction Cimeron River on highway 23, 25 June 1992, E.G. Riley (1^o, TAMU). NEW MEXICO: Eddy Co., 26 miles east Carlsbad, 2 June 1977 (1^Q, TAMU), same, 3 June 1977, malaise trap (West) (1^Q, TAMU), same, 9 June 1977, grasses, plot W 20, 21, 26, 27, plant #80 (1³, TAMU). TEXAS: Brewster Co., Big Bend National Park, July 1973 (7Å, 3♀, LSAM), Big Bend National Park, North Rosillos Mountains, Buttrill Spring, malaise trap, 10-14 July 1991 R. Vogtsberger (1^Q, TAMU), Hidalgo Co., Lower Rio Grande Valley National Wildlife Refuge, McManus unit, 26.05380°N, 98.04987°W, 3 September 2008, UV light, J. King and E. Riley, 22 primary forest (1♀, TAMU), Llano Co., Tow, 21 March 1982, W.F. Chamberlain (1♂, TAMU), Uvalde Co., Garner State Park, elevation 1400', 21 July 1986, 86/017, J.B. Woolley and G. Zolnerowich (11^Q, TAMU). UTAH: Washington Co., 29 April 1938, Christenson, No. 12424 (1⁽²⁾, USNM). **MEXICO**: Puebla, 4.7 miles southwest La Cumbre, 23 July 1987, 5100', J.B. Woolley and G. Zolnerwich, 87/055 (1³, TAMU), Zacatecas, 4 miles northeast Concepcion del Oro, 4 July 1984, J.B. Woolley 84/014 (2♂, 1♀, TAMU).

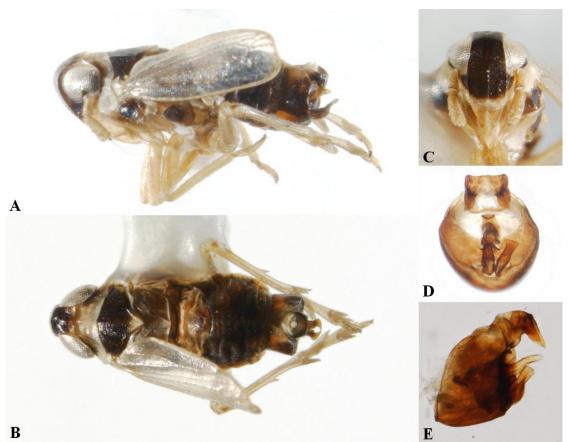


Figure 11. Features of *Flavoclypeus latidens*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.7 Flavoclypeus nigrifacies (Muir, 1918), new comb.

Delphacodes nigrifacies Muir, 1918: 428.

Delphacodes xerosa Caldwell, 1951; synonymy by Kennedy et al., 2012: 405.

Type locality. Martinique, Fort de France.

Diagnosis. General body color glossy dark brown, with white to yellow clypeus,

second antennal segment, posterior edge of pronotum, and legs; wings clear. Length

1.41-2.67 mm, varied by sex and wing morph. Parameres flattened, broadest apically, with elongate outer angles. Aedeagus tubular, bearing several rows of retrose spines.Segment 10 bearing pair of broad, truncate processes.

Color. Frons glossy dark brown with contrasting yellow clypeus; carinae concolorous. First antennal segment brown, second segment yellow. Vertex dark yellow to light brown. Prothorax dark brown with paler posterior margin and sometimes with paler lateral margins; mesothorax dark brown with yellow scutellum; tegulae and adjacent lateral edges of mesothorax yellow. Coxae dark brown, legs yellow; wings hyaline, veins indistinct. Abdomen fading from brown anteriorly to yellow posteriorly; posterior edge of each abdominal segment darker than anterior edge. Sexually dimorphic coloration with females typically similar in coloration to males, or with paler variations such as yellow to orange vertex and pronotum.

Structure. Length 3° macropter: 2.24 mm (n = 1); 2° macropter: 2.58 mm (2.49-2.67, n = 2). Length 3° brachypter: 1.53 mm (1.41-1.65, n = 10); 2° brachypter: 1.70 mm (1.53-1.80, n = 14).

Head. Frons approximately twice as long as broad (l:w 1.83:1); carinae strongly evident. Frontal median carina forked just below fastigium. Vertex just wider than long (l:w 0.94:1). Antennal segment I slightly longer than wide (l:w 1.14:1); second antennal segment approximately twice as long as first (I:II 0.50:1).

Thorax. Mesonotum about twice as long of pronotum (pronotum 1:mesonotum 1 0.55:1). Median carina of mesonotum becoming obsolete on scutellum; lateral carinae of pronotum and mesonotum diverging, curved posteriorly, not reaching hind margin. Wings vary in length, leaving abdominal segments 7-10 exposed in some brachypters and only the pygofer in others; wings rounded at apex. Acuminate calcar bearing row of 15-20 (n = 5) fine, black-tipped teeth on outer margin.

Abdomen. Pygofer approximately triangular in lateral view, much wider ventrally than dorsally. Diaphragm well-developed, armature concave, wider than tall, with lateral dorsally-directed projections. Parameres broad, reaching broadest point apically, outer angles elongate, pointing laterally; inner angles acute, pointing medially; basal angles strongly evident. Aedeagus tubular, broadest basally, bearing several irregular rows of retrose spines. Segment 10 bearing a pair of caudally-projected processes, truncate at apices.

Hosts.

Chamaecrista fasciculata (Michx.) Greene (partridge pea) (Kennedy et al. 2012) *Cynodon dactylon* Pers. (Bermudagrass) (Calvert et al. 1987) *Paspalum notatum* Flueggé (bahiagrass) (Kennedy et al. 2012)
Poaceae (reported as Gramineae grass) (Ballou 1936) *Stenotaphrum secundatum* Kuntze (St. Augustine grass) (Calvert et al. 1987)

Distribution. USA (FL), Belize, Bolivia, Colombia, Costa Rica, Ecuador, Grenada, Guyana, Jamaica, Martinique, Mexico, Panama, Puerto Rico, St. Thomas, St. Vincent and the Grenadines, Venezuela. Also reported from Dominica (Fennah 1959), Montserrat (Fennah 1959), St. Lucia (Fennah 1959).

Etymology. Muir (1918) did not specify the etymological origin, but the specific name is presumably formed from the Latin adjective *niger* meaning "black" and the Latin noun *facies* meaning "face", in reference to the dark frons.

Remarks. Muir and Giffard (1924) note that the type of *L. andromeda* Van Duzee from Demerara, British Guinea (R.J. Crew, April 2, 1901) is actually *D. nigrifacies*. These two species are very similar externally; *F. nigrifacies* has only one pair of processes on segment 10 instead of 2 as observed in *F. andromedus*.

At the outset of this project, *Delphacodes xerosa* was considered a separate taxon, but it was synonymized with *F. nigrifacies* during the duration of this project based on comparison of the type specimens (Kennedy et al. 2012).

Type material examined. MARTINIQUE: Holotype (male brachypter, AMNH): "Fort de France / Martinique, W.I. / June 27, 1911 // [red paper] TYPE OF / D. nigrifacies / Muir // Am. Mus. Nat. Hist. / Dept. Invert. Zool. / No. 24254 // [red paper] HOLOTYPE / DELPHACODES / NIGRIFACIES / MUIR". Other material examined. See Appendix A.

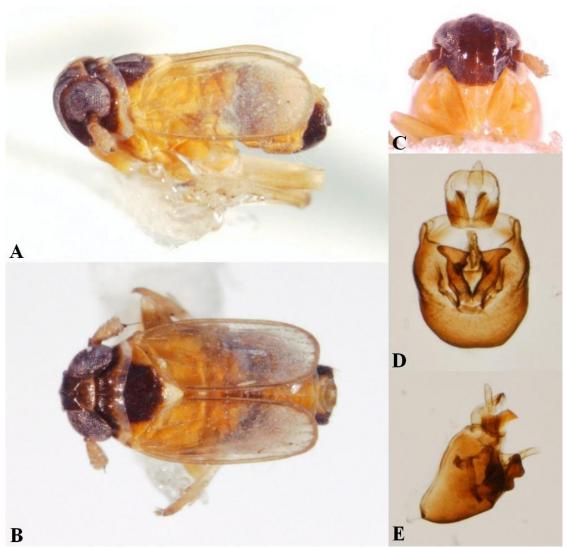


Figure 12. Features of *Flavoclypeus nigrifacies*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.8 Flavoclypeus nigriscutellatus (Beamer 1947), new comb.

Delphacodes nigriscutellata Beamer 1947: 62.

Caenodelphax nigriscutellata (Beamer 1947), combination by Bouchard et al. 2002: 49.

Type locality. US, Kansas: Douglas Co.

Diagnosis. General body color glossy dark brown, with white to yellow clypeus, antennae, pronotum, and legs; wings clear. Length 1.70-3.26 mm. Parameres broad, inner angles elongate. Aedeagus tapering from broad base to narrow apex, incurved ventrally in apical third, bearing about 4 retrose teeth dorsally and 2 retrose teeth on both lateral margins. Segment 10 bearing pair of elongate, pointed, caudally projected processes.

Color. General body color dark brown to black, carinae concolorous with body. Posterior edge of pronotum white to yellow; in some specimens, entire pronotum white to yellow, in contrast with dark brown mesonotum, or white to yellow with darker brown patches on lateral margins. Vertex typically dark brown but appearing paler yellow in some specimens. First and second antennal segments, clypeus, tegulae, scutellum, and legs yellow. Wings translucent, veins darker. Pygofer brown. Sexually dimorphic coloration with females typically paler, uniform white to yellow.

Structure. Length \Im macropter: 3.26 mm (n = 1); \Im macropter: none observed. Length \Im brachypter: 1.70 mm (n = 1); \Im brachypter: none observed. *Head.* Head, including eyes, approximately equal in width to pronotum. Frons longer than wide (1:w 1.39:1). Frons widest at middle, between ocelli; slightly narrower at base than at apex. Median carina of frons forked below fastigium. Vertex length approximately equal to width (1:w 1.04:1). Antennal segment I equal in length and width (1:w 1:1); second antennal segment approximately twice as long as first (I:II 0.43:1).

Thorax. Mesonotum not quite twice as long as pronotum (pronotum l:mesonotum l 0.60:1); pronotal carinae evident, lateral mesonotal carinae evident, median mesonotal carina becoming obsolete before scutellum. Wings (brachypter) longer than wide. Wings (macropter) extend beyond base of abdomen by one-third. Wings rounded at apex in both forms. Calcar bearing continuous row of 14-16 (n = 2) black-tipped teeth.

Abdomen. Pygofer approximately ovular in lateral view, slightly wider ventrally than dorsally; anterior margin longer than posterior margin. Parameres with elongate, medially-projected inner angles, much shorter laterally-projected outer angles, and short caudally-projecting processes at base. Aedeagus broadest basally, thinnest apically, incurved ventrally towards apex, bearing row of about 4 retrose teeth along dorsal margin and a pair of retrose teeth on both lateral margins at the base. Processes of segment 10 long, slender, caudally projected, tapering to pointed apex. Segment 11 elongate, roughly as long as height of segment 10.

Hosts.

Andropogon gerardii Vitman (big bluestem; reported as A. furcatus Muhl; see remarks) (Beamer 1947)
Eleocharis compressa Sull. (flatstem spikerush) (Bouchard et al. 2002)
Eleocharis elliptica Kunth (elliptic spikerush) (Bouchard et al. 2002)
Spartina pectinata Bosc ex Link (prairie cordgrass, UDCC)
Sporobolus heterolepis (A. Gray) A. Gray (prairie dropseed; see remarks) (Bouchard 1997)

Distribution. USA: IA, KS, WI. Also reported from USA (MN, SD) (Bouchard 1998) and Canada (AB, MB, ON) (Bouchard 1997).

Etymology. Beamer (1947) did not specify the origin of this name, but it was presumably formed from the Latin adjective *niger* meaning "black" and the Latin noun *scutellum*, diminutive of *scutum*, meaning "shield", in reference to the dark scutellum.

Remarks. In his original diagnosis, Beamer (1947) described *D. nigriscutellata* as being similar to *D. shermani* but with differences in coloration. In addition to the color differences described by Beamer, there are distinctions in structure between these

species, such as *D. shermani*'s avicephaliform parameres, wide, rectangular aedeagus, greater body length to width ratio, and longer frons. For these reasons, *D. shermani* was excluded from the ingroup.

Beamer (1947) reports that "this species was collected in Douglas Co., Kans., by sweeping around the edge of a marsh which had a fair stand of *Andropogon furcatus* Muhl. It was not taken in any other stand of this grass although several other locations were swept." *Sporobolus heterolepis* (A. Gray) A. Gray (prairie dropseed) was additionally listed as a potential host by Bouchard (1997).

Bouchard (1998) remarks that this species is very rare and occurs in low abundance, necessitating intensive collecting efforts. It has been collected from April through September. Wallner (2010) established that *C. nigriscutellatus* is a prairie habitat specialist and intolerant of prairie degradation.

Type material examined. USA: KANSAS: Plesiotype/holomorphotype (male macropter, SEMC): "Meade Co. Kans / 9-13 1944 / R. H. Beamer // [handwritten, orange paper] Holomorphotype / Delphacodes / nigriscutellata / R.H. Beamer". Allomorphotype (female macropter, SEMC): "Douglas Co. Kans / Apr. 18 1946 (4) / R.H. Beamer // [handwritten, orange paper] Allomorphotype / Delphacodes / nigriscutellata / R. H. Beamer". Paratype (male brachypter, SEMC), "Douglas Co. Kans / 4-12-1946 (4) / R. H. Beamer // [blue paper] PARATYPE / Delphacodes / nigriscutellata / R.H. Beamer", paratype (male brachypter, AMNH): "Douglas Co. Ks / Apr. 18 1946 (4) / R. H. Beamer", paratype (male brachypter, AMNH): "Douglas Co. Ks

nigriscutellata / R.H. Beamer", paratype (female brachypter, AMNH): "Douglas Co. Ks / Apr. 18 1946 (4) / R.H. Beamer // [blue paper] PARATYPE / Delphacodes / nigriscutellata / R.H. Beamer".

Other material examined. USA: IOWA: <u>Story Co.</u>, Ames exp. sta., 6 August 1897 (1승, USNM). WISCONSIN: <u>Jefferson Co.</u>, Faville Prairie State Natural Area, N43.14646 W88.87928, 22 August 2005, A.M. Wallner, vacuum from prairie cordgrass (3승, UDCC).

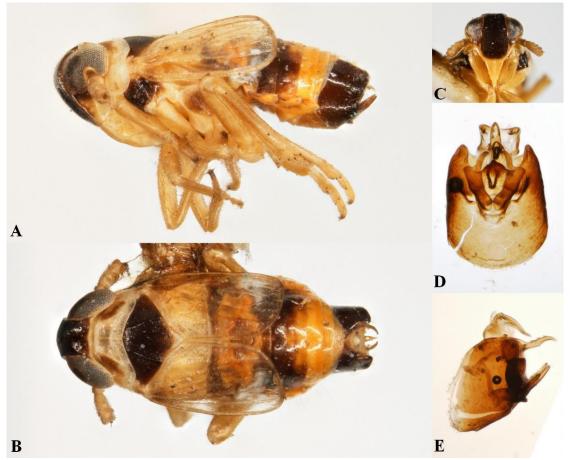


Figure 13. Features of *Flavoclypeus nigriscutellatus*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.2.9 Flavoclypeus nitens (Muir and Giffard, 1924), new comb.

Delphacodes nitens Muir and Giffard, 1924: 27.

Type locality. US, Ohio: Columbus.

Diagnosis. General body color glossy dark brown to black with yellow to orange clypeus, antennae, and legs; wings dark. Length 2.13-3.03 mm, with females larger. Parameres broad, constricted most narrowly subapically, with outer angles longer than inner angles. Aedeagus thick, bearing row of about 6 teeth on both lateral margins, third row of about 4 teeth on dorsal margin. Segment 10 bearing pair of pointed, slender processes, curved ventrally.

Color. General body color dark brown to black; glossy. Carinae concolorous with body. Clypeus, first and second antennal segments, legs, and scutellum pale yellow to orange. Posterior compartments of vertex lighter brown than areolet, anterior compartments of vertex, and pronotum. Wings dark brown, veins concolorous. Sexually dimorphic coloration with females typically paler, uniform white to yellow.

Structure. Macropters: none observed. Length 3° brachypter: 2.35 mm (2.13-2.46, n = 10); length 9° brachypter: 2.84 mm (2.62-3.03, n = 7).

Head. Head slightly narrower than pronotum. Frons twice as long as broad (l:w 1.94:1), widest at middle between ocelli; carinae strongly evident. Median carina of frons forked below fastigium. Vertex longer than wide (l:w 1.17:1); carinae evident. Antennal segment I slightly longer than wide (l:w 1.22:1); second antennal segment approximately twice as long as first (I:II 0.46:1); second segment bearing sensory fields approximately arranged in rows.

Thorax. Mesonotum not quite twice as long as pronotum (pronotum l:mesonotum l 0.62:1). Median carina of mesonotum becoming obsolete on scutellum; lateral carinae diverging posteriorly to reach hind margin. Wings rounded apically. Calcar bearing continuous row of 14-17 (n = 5) very fine black-tipped teeth.

Abdomen. Pygofer approximately quadrate in lateral view, just wider ventrally than dorsally; in caudal view, opening roughly quadrate, slightly wider than tall. Diaphragm well-developed; armature projecting caudally. Parameres broad basally, constricted most narrowly distally before broadening apically; basal angles barely evident, inner angles subacute, outer angles rounded, longer than inner angles; apex truncate. Suspensorium ring-shaped. Aedeagus thick, tubular, broadest basally, orifice at apex. Two rows of approximately 6 teeth on opposite lateral margins of aedeagus, spanning from mid-length to apex; third row of approximately 4 teeth located dorsally. Segment 10 with a pair of pointed, slender processes, slightly broader basally, bent at right angle to project ventrally. Segment 11 elongate, roughly equal in length to height of segment 10, slightly pointed at apex.

Hosts. None reported.

Distribution. USA (DC, DE, IL, MD, NC, TN, TX), Mexico. Also reported from USA (OH) (Muir and Giffard 1924).

Etymology. The specific name is presumably formed from the Latin *niteo* meaning "shine" in reference to the glossy quality of the habitus. Muir and Giffard (1924) did not specify the origin but refer to this species as "shiny black".

Remarks. As noted above, *F. nitens* is superficially similar to *F. atridorsum* due to their very dark coloration, including an entirely dark pronotum and dark wings; however, *F. nitens* is larger, with the male brachypter more than half a millimeter longer on average, and distributed in the eastern half of the United States whereas *F. atridorsum* is only reported from Oregon.

This species has been collected from April through September.

Type material examined. USA: MARYLAND: Paratype (male brachypter, USNM): "Plummers I / May-9-13 Md / WLMcAtee / Collector // [orange paper] Paratype", paratype (male brachypter, USNM): "Plummers I / May-18-13 Md / WLMcAtee / Collector // [orange paper] Paratype".

Other material examined. See Appendix A.

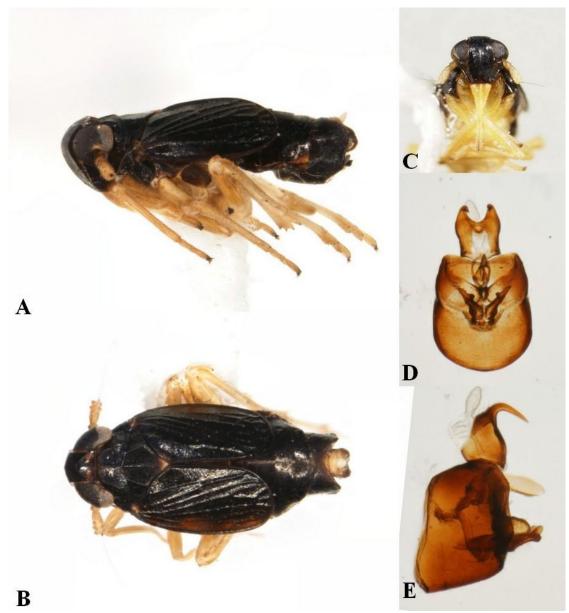


Figure 14. Features of *Flavoclypeus nitens*. A. Lateral habitus, B. Dorsal habitus, C. Frons, D. Pygofer, caudal view, E. Pygofer, lateral view.

2.3 Taxa Excluded from *Caenodelphax* and *Flavoclypeus*

The following taxa were excluded from both *Caenodelphax* and *Flavoclypeus* following this morphological investigation: Delphacodes aterrima, D. balli, D. livida, D. recurvata, D. shermani, and D. sucinea. These species were excluded on the basis of marked differences in structure and color as well as the results of the phylogenetic analysis; the tree in Figure 5 clearly illustrates that they are not part of a monophyletic grouping with the species described above. D. aterrima is distinguished by not bearing any processes on segment 10, in sharp contrast to the other species examined. D. aterrima, D. recurvata, and D. sucinea display a tall, narrow, approximately rectangular pygofer in lateral view, unlike the wide quadrate or triangular pygofer shapes observed in the ingroup. D. balli is marked by a greatly expanded dorsocaudal margin of the pygofer, and D. balli, D. livida, and D. recurvata have the frons and clypeus concolorous instead of contrasting. D. recurvata bears distinctly bilobed parameters unlike any seen in the ingroup, and D. recurvata and D. sucinea both bear processes on the aedeagus that are substantially more elongate than any teeth or spines observed in the ingroup species' aedeagi. The length-to-width ratio of the first antennal segment in *D. sucinea* is much greater than those of the other examined species. D. sucinea's general body color is a glossy honey color instead of dark brown to black, and there is a distinct white stripe at the apices of the elytra; similarly, the general body color of *D. recurvata* and *D. shermani* is tan as opposed to dark brown or black, and the genae of *D. shermani* are darker than the frons, which is not observed in any ingroup species. Additionally, the frons of *D. shermani* is longer and the body length-to-width ratio is higher than observed in the ingroup species. These morphological differences are greater than would be expected due to normal intrageneric variation, although molecular data, when it becomes available, will assist in confirming their phylogenetic relationships. As noted, these species do not belong in *Delphacodes* but their proper placement remains undetermined at this time.

2.4 Discussion

This revision restricted *Caenodelphax* to a monotypic genus consisting of the type species, *C. teapae*. This definition corresponds largely with the original generic definition established by Fennah (1965), who described *Caenodelphax* as a strictly Neotropical genus. Although Fennah described it as comprising two species, and Hamilton broadened it to encompass two additional species, the synonymization of *C. philyra* and *C. teapae* and the transfer of *F. atridorsum* and *F. nigriscutellatus* to *Flavoclypeus* hereby reduce it to only one species. Despite the reduction in number of taxa included in *Caenodelphax*, it remains a widespread and economically important genus due to the expansive feeding habits of the type species (Table 6).

Results from the phylogenetic analysis suggest that *Delphacodes livida* and *D*. *balli* may be closely allied with *C. teapae*, but their relationship is uncertain at this time. Both *D. balli* and *D. livida* exhibit an expanded dorsocaudal margin of the pygofer, which is not observed in *C. teapae*; *C. teapae* also displays serrate apices of the processes on segment 10, compared to smooth apices in the other two species.

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These features are usually considered generic-level, suggesting that these three species do not belong to the same genus. Additional phylogenetic analyses with expanded taxon sampling, and ideally incorporating molecular data, are needed to determine whether *D. livida* and *D. balli* should be assigned to *Caenodelphax*.

The new genus *Flavoclypeus* comprises eight former *Delphacodes* species considered but ultimately rejected for inclusion in *Caenodelphax*. *Flavoclypeus* is a widespread New World genus, ranging as far south as Bolivia and Brazil (*F. nigrifacies*) and as far north as Canada (*F. incurvus*, *F. nigriscutellatus*), from the Pacific coast (*F. atridorsum*) to the Atlantic (e.g., *F. andromedus*, *F. nitens*) and the Caribbean islands (*F. andromedus*, *F. nigrifacies*). While host plant records exist for some species (Table 6), suggesting that this genus feeds primarily on grasses and sedges, other species' feeding habits remain unknown.

Future work on this genus should incorporate molecular analyses to provide a better-informed understanding of its interrelationships. Better-targeted primers for PCR are essential for yielding meaningful COI data for phylogenetic analysis. A key to the females of this genus is still lacking, and greater collection efforts will likely increase the known distribution significantly.

Flavoclypeus			
Species	Hosts	Common	Source
		name	
Caenodelphax	Axonopus compressus (Sw.) P. Beauv.	broadleaf	Fennah
teapae		carpetgrass	1959
	Crotalaria L.	rattlebox	Leonard
			1933

Table 6. Summary of recorded host plants for species of *Caenodelphax* and *Flavoclypeus*

	Cucurbita maxima Duchesne	winter squash	Label data
	<i>Cymbopogon citratus</i> (D.C. ex Nees) Stapf	lemon grass	Wolcott 1923
	Cynodon dactylon (L.) Pers.	Bermudagrass	Label data
	Daucus L.	carrot	Wolcott 1923
	Paspalum notatum Flueggé	bahiagrass	Label data
	Phaseolus vulgaris L.	kidney bean	Label data
	Saccharum L.	sugarcane	Wolcott 1923
	Solenostemon scutellarioides (L.) Codd	common coleus	Ballou 1936
	Urochloa plantaginea (Link) R. Webster	plantain signalgrass	Wilson 2005
Flavoclypeus andromedus	Paspalum L.	crowngrass	Osborn 1926
	Eleocharis R. Br	spikerush	Label data
Flavoclypeus	Setaria texana W.H.P. Emery	Texas	Wilson et
latidens		bristlegrass	al. 1994
Flavoclypeus nigrifacies	Chamaecrista fasciculata (Michx.) Greene	partridge pea	Kennedy et al. 2012
	Cynodon dactylon Pers.	Bermudagrass	Calvert et al. 1987
	Paspalum notatum Flueggé	bahiagrass	Kennedy et al. 2012
	Poaceae (reported as Gramineae grass)	St Augustine	Ballou 1936 Calvert et
	Stenotaphrum secundatum Kuntze	St. Augustine grass	al. 1987
Flavoclypeus nigriscutellatus	Andropogon gerardii Vitman	big bluestem	Beamer 1947
	Eleocharis compressa Sull.	flatstem spikerush	Bouchard et al. 2002
	Eleocharis elliptica Kunth	elliptic spikerush	Bouchard et al. 2002
	<i>Sporobolus heterolepis</i> (A. Gray) A. Gray	prairie dropseed	Bouchard 1997

REFERENCES

- Arnett, R. H., Jr., G. A. Samuelson, and G. M. Nishida. 1993. The Insect and Spider Collections of the World, 2nd ed. Sandhill Crane Press, Gainesville, Florida. 310 pp.
- Asche, M. 1985. Zur Phylogenie der Delphacidae Leach, 1815 (Homoptera: Cicadina: Fulgoromorpha). Marburger Entomologische Publikationen Marburger Entomologische Publikationen 12(1):1-398 and 2(2): 399-910.
- Asche, M. 1990. Vizcayinae, a new subfamily of Delphacidae with revision of *Vizcaya* Muir (Homoptera: Fulgoroidea) a significant phylogenetic link. Bishop Museum Occasional Papers 30: 154-187.
- **Ballou, C.H.** 1936. Insect notes from Costa Rica in 1935. Insect Pest Survey Bulletin 16: 473.
- Beamer, R.H. 1947. Some new species of *Delphacodes* (continued) with two old ones (Homoptera: Fulgoridae). Journal of the Kansas Entomological Society 20(2): 58-71.
- **Beamer, R.H.** 1948a. Some new species of Delphacodes (continued), Part III. Journal of the Kansas Entomological Society 21(1): 1-10.
- Beamer, R.H. 1948b. Some new species of Delphacodes (Homoptera: Fulgoridae: Delphacinae), Part IV. Journal of the Kansas Entomological Society 21(3): 96-110.

- Bouchard, P. 1997. Insect diversity of four alvar sites on Manitoulin Island, Ontario. Unpublished M. Sc. thesis. McGill University, Montreal. 93 pp.
- **Bouchard, P.** 1998. Insect diversity in alvars of southern Ontario. Technical Report to Federation of Ontario Naturalists, Don Mills, Ontario. 87 pp.
- Bouchard, P., K.G.A. Hamilton and T.A. Wheeler. 2002. Diversity and conservation status of prairie endemic Auchenorrhyncha(Homoptera) in alvars of the Great Lakes region. Proceedings of the Entomological Society of Ontario 132: 39-56.
- **Caldwell, J.S. and L.F. Martorell.** 1951 [dated1950]. Review of the Auchenorynchous [sic] Homoptera of Puerto Rico. Part II. The Fulgoroidea except Kinnaridae. Journal of Agriculture of the University of Puerto Rico 34(2): 133-269.
- **Crawford, D.** 1914. A contribution toward a monograph of the homopterous insects of the family Delphacidae of North and South America. Proceedings of the United States National Museum 46: 557-640.
- Cronin, J.T., K.J. Haynes, and F. Dillemuth. 2004. Spider effects on planthopper mortality, dispersal and spatial population dynamics. Ecology 85: 2134-2143.
- **Denno, R.F., M.S. Mitter, G.A. Langellotto, C. Gratton, and D.L. Finke.** 2004. Interactions between a hunting spider and a web-builder: consequences of

intraguild predation and cannibalism for prey suppression. Ecological Entomology 29(5): 566-577.

- **Dijkstra, E., J.M. Rubio, and R.J. Post.** 2003. Resolving relationships over a wide taxonomic range in Delphacidae (Homoptera) using the COI gene. Systematic Entomology 28(1): 89-100.
- **Farris, J. S.** 1969. A successive approximations approach to character weighting. Systematic Zoology 18(4): 374-385.
- Felsenstein, J. 1985. Confidence limits on phylogenies: An approach using the bootstrap. Evolution 39: 783-791.
- Fieber, F.X. 1866. Grundzüge zur generischen Theilung der Delphacini. Verhandlungen der Kaiserlich-Königlichen Zoologish-botanische Gesellschaft in Wien 16: 517-534.
- **Fennah, R.G.** 1959. Delphacidae from the Lesser Antilles (Homoptera: Fulgoroidea). Bulletin of the British Museum (Natural History), Entomology 8: 245-265.
- **Fennah, R.G**. 1965. New Species of Fulgoroidea (Homoptera) from the West Indies. Transactions of the Royal Entomological Society, London 117(4): 95-126.
- Fennah, R.G. 1967. Fulgoroidea from the Galapagos Archipelago. Proceedings of the California Academy of Sciences 35(4): 53-102.

Fowler, W.W. 1904. Order Rhynchota. Suborder Hemiptera-Homoptera.

(Continued). Biologia Centrali-Americana; or, Contributions to the knowledge of the fauna and flora of Mexico and Central America 1: 77-124.

- **Fowler W.W.** 1905. Order Rhynchota. Suborder Hemiptera-Homoptera. (Continued). Biologia Centrali-Americana; contributions to the knowledge of the fauna and flora of Mexico and Central America 1: 125-139.
- Hamilton, K.G.A. 2002. Homoptera (Insecta) in Pacific Northwest grasslands. Part 1-New and revised taxa of leafhoppers and planthoppers (Cicadellidae and Delphacidae). Journal of the Entomological Society of British Columbia 99: 3-31.
- International Commission on Zoological Nomenclature (ICZN). 1961. Opinion 602. *Delphax* Fabricius, 1798 (Insecta, Hemiptera); Interpretation under the plenary powers. Bulletin of Zoological Nomenclature 18(4): 246-248.
- International Commission on Zoological Nomenclature (ICZN). 1999. International Code of Zoological Nomenclature, Fourth Edition. International Trust for Zoological Nomenclature, 306 pp.
- Kennedy, A.C., C.R. Bartlett, and S.W. Wilson. 2012. An annotated checklist of the delphacid planthoppers (Hemiptera: Delphacidae) of Florida with the description of three new species and the new genus *Meristopsis*. Florida Entomologist 95(2): 394-420.

- **Kirkaldy, G.W.** 1904. Some new Oahuan (Hawaiian) Hemiptera. Entomologist 37: 174-179.
- Liu, X.D., X.X. Zhang, H.F. Guo, and Y.J. Luo. 2001. Control function of spider community to planthopper in the rice field. Acta Ecologica Sinica 21(1): 100-105.
- Maw, H.E.L., R.G. Foottit, K.G.A. Hamilton, and G.G.E. Scudder. 2000. Checklist of the Hemiptera of Canada and Alaska. NRC Research Press, Ottawa. read online: http://www.ccebook.org/preview/0660181657/Checklistof-the-hemiptera-of-Canada-and-Alaska.
- Muir, F.A.G. 1918. Homopterous notes II. Proceedings of the Hawaiian Entomological Society 3: 414-429.
- Muir, F.A.G. and Giffard, W.M. 1924. Studies in North American Delphacidae. Bulletin of the Experiment Station of the Hawaiian Sugar Planters' Association, Entomological series no. 15: 1-53.
- **Osborn, H.** 1926. Faunistic and ecological notes on some Cuban Homoptera. Scientific Contributions series of the Tropical Plant Research Foundation 2: 335-366; plates 30-31.
- **Osborn, H.** 1929. Notes on Porto Rican Homoptera. Journal of the Department of Agriculture of Porto Rico 13: 81-112.

- **Osborn, H.** 1935. Survey of Porto Rico and the Virgin Islands Homoptera (excluding Sternorhynchi). Scientific Survey of Porto Rico and the Virgin Islands 14: 111-260.
- **Osborn, H.** 1938. The Fulgoridae of Ohio. Bulletin of the Ohio Biological Survey 6: 283-349.
- Page, R.D.M. 1996. TreeView: An application to display phylogenetic trees on personal computers. Computer Applications in the Biosciences 12(4): 357-358.
- Poe, S. and J.J. Wiens. 2000. Character selection and the methodology of morphological phylogenetics, pp. 20-36 in J.J. Wiens (ed.), Phylogenetic Analysis of Morphological Data. Smithsonian Institution Press, Washington, D.C.
- **Ronquist, F. and J.P. Huelsenbeck.** 2003. MRBAYES 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19: 1572-1574.
- Simon, C., F. Frati, A. Beckenbach, B. Crespi, H. Liu, and P. Flook. 1994. Evolution, Weighting, and Phylogenetic Utility of Mitochondrial Gene Sequences and a Compilation of Conserved Polymerase Chain Reaction Primers. *Annals of the Entomological Society of America*. 67(6): 651-701.

- Swofford, D.L. 1998. PAUP*. Phylogenetic Analysis Using Parsimony (*and Other Methods). Version 4. Sinauer Associates, Sunderland, Massachusetts.
- Urban, J.M., Bartlett, C.R., and J.R. Cryan. 2010. Evolution of Delphacidae (Hemiptera: Fulgoroidea): combined-evidence phylogenetics reveals importance of grass host shifts. Systematic Entomology 35: 678-691.
- Urban, J.M., and J.R. Cryan. 2012. Two ancient bacterial endosymbionts have coevolved with the planthoppers (Insecta: Hemiptera: Fulgoroidea). BMC Evolutionary Biology 12(1): 87.
- USDA, NRCS. 2012. The PLANTS Database (http://plants.usda.gov, 21 December 2012). National Plant Data Team, Greensboro, NC 27401-4901 USA.
- Van Duzee, E.P. 1907. Notes on Jamaican Hemiptera: A report on a collection of Hemiptera made on the island of Jamaica in the spring of 1906. Bulletin of the Buffalo Society of Natural Sciences 8(5): 3-79.
- Van Duzee, E.P. 1917. Catalogue of the Hemiptera of America North of Mexico (excepting the Aphididae, Coccidae and Aleurodidae). University of California Publications, Technical Bulletins, vol. 2. University of California Press, Berkeley, pp. i-xiv, 1-902.
- Wagner, W. 1963 [dated 1962]. Dynamische Taxonomie, angewandtauf die Delphaciden Mitteleuropas. Mitteilungenausdem Hamburgischenzoologischen Museum und Institut 60: 111-180.

- Wallner, A. 2010. Evaluating North American tallgrass prairie quality using the Auchenorrhyncha quality index. Doctoral Dissertation. University of Illinois at Urbana-Champaign. Urbana, Illinois.
- Wilson, S.W. 2005. Keys to the families of Fulgoromorpha with emphasis on planthoppers of potential economic importance in the southeastern United States (Hemiptera: Auchenorrhyncha). Florida Entomologist 88(4): 464-481.
- Wilson, S.W., C. Mitter, R.F. Denno, and M.R. Wilson. 1994. Evolutionary patterns of host plant use by delphacid planthoppers and their relatives, pp. 7–113 in R. F. Denno and T. J. Perfect (eds.), Planthoppers: Their Ecology and Management. Chapman & Hall, New York.
- Wilson, S.W. and A.G. Wheeler, Jr. 2010. Planthopper (Hemiptera: Fulgoroidea) diversity of weeping lovegrass (*Eragrostis curvula*), an introduced host of littleknown, rarely collected native species. Entomologica Americana 116(3/4): 98–106.
- Wolcott, G.N. 1923. Insectae Portoicensis. A preliminary annotated check-list of the insects of Porto Rico, with descriptions of some new species. Journal of the Department of Agriculture Porto Rico 7(1): 1-313.
- **Wolcott, G.N.** 1950. The insects of Puerto Rico. [dated 1948]. Journal of Agriculture of the University of Puerto Rico 32(1): 1-224.

APPENDIX A

MATERIAL EXAMINED

Caenodelphax teapae

USA: FLORIDA: Broward Co., Fort Lauderdale, Lauderhill, Invarray, 24 July 1999, C. R. Bartlett (7⁽²⁾, UDCC), 5 July 2000, C. R. Bartlett, sweep, lawn/sedge in shallow ditch (3⁽²⁾, UDCC). ANTIGUA AND BARBUDA: Antigua, St. John's, June 1962, J. Maldonado G. (1⁽²⁾, USNM). **BELIZE**: <u>Cayo District</u>, near Teakettle Bank, Pook's Hill, 4-7 January 2003, C. R. Bartlett, sweeping $(8^{\uparrow}_{\circ}, 4^{\bigcirc}_{+}, \text{UDCC})$, 2 July 2003, C.R. Bartlett (1Å, UDCC). **BOLIVIA**: Santa Cruz Dept., 3.7 km SSE Buena Vista, Hotel Flora y Fauna, 430m 17°29'S 63°33'W, MC Thomas, 14-28 October 2000 (1^Q, UDCC). BRAZIL: Piracicaba, Sao Paolo, 9 December 1964, C. A. and W.E. Triplehorn, blacklight trap (1⁽²⁾, UDCC), Rondonia, 8 km. N. Porto Velho, 7 October 1984, J.F. Cornell collection (2³, UDCC). **COLOMBIA**: Putumayo, PNN La Paya Cabana La Paya, 0°2'S 75°12'W, 330 m, 24-25 September 2001, Pantrap, D. Campos (2Å, UDCC). **COSTA RICA**: Alajuela Province, Cano Negro, at dock path, A.E.Z. Short, 14 January 2004, Cartago Province, Turrialba, 600-700 m., 12 August 1975, N.L.H. Krauss (1⁽²⁾, USNM), Cartago Province, Tapanti National Park, Arboles Caidos Trail, 16 July 2011, sweep, A. Kennedy (1⁽²⁾, UDCC), Tapanti National Park,

road by visitor center, 18 July 2011, sweep, A. Kennedy (1⁽²⁾, UDCC), Heredia Province, near Puerto Viejo, La Selva Biological Station, station grounds, 23 February-2 March 2004, C. R. Bartlett, J. Cryan, J. Urban (9⁽²⁾, UDCC), 10°26'N, 84°00'W, 21 March 2005, S.M. Clark (1, 1, 1, BYUC), Limón Province, Pandora, 150 feet, 22 August 1963, S.L.W. (2Å, BYUC), Puntarenas Province, Rincon, Osa Peninsula, 100 feet, 11 August 1966, S.L.W. (1, BYUC). CUBA: Havana Province, Hoya Colorado, 23 August 1917, Harold Morrison (1Å, USNM), Caimito, 23 August 1917, Harold Morrison (1², USNM). **DOMINICA**: St. George Parish, Roseau, November 1967, N.L.H. Krauss (13° , 19° , USNM), St. Joseph Parish, July 1963, J. Maldonado C. (1⁽²⁾, USNM), Springfield Estate, malaise in humid forest, 15-20 March 2003, M.E. Irwin, M.B. Shephard, E. Benson, G. Carner (1⁽²⁾, UDCC), Sylvania, November 1967, N.L.H. Krauss (2^Q, USNM), St. Paul Parish, Pont Casse, J. Maldonado C. (2Å, USNM). **DOMINICAN REPUBLIC**: La Vega Province, 6 mi NW or Route 1 on road to Constanza, 27 June 1998, blacklight, R.E. Woodruff, R.M. Baranowski (1^Q, UDCC), Constanza, 1 August 1978, R.O. Schuster and R.S. Rominger (2⁽²⁾, CDAE), María Trinidad Sánchez Province, Cabrera, 1 August 1978, R.O. Schuster and R.S. Rominger (1⁽²⁾, CDAE), Samaná Province, Playa Rincon, 31 July 1978, R.O. Schuster (1Å, CDAE). ECUADOR: Provincia de Francisco de Orellana, Yasuni National Park, 29 April 2005, C.R. Bartlett, N. Nazdrowicz, D. Chang (13, UDCC), Santo Domingo de los Colorados, 6 March 1973, M.A. Deyrup (1⁽¹), UDCC). **EL SALVADOR**: <u>La Libertad</u>, Quezaltepeque, 21 June 1961, M.E. Irwin (13, CDAE), 4 August 1963, D. Cavagnaro and M.E. Irwin (23, CDAE), San

Salvador, October 1965, N.L.H. Krauss (1Å, 1º, USNM). FRENCH GUIANA: 8 kilometers west of Risquetout, 45 meters, 10-11 June 2005, J.E. Eger, N04 55.097, W052 33.121 (2 $\cancel{2}$, UDCC). **GRENADA**: Aug Busck (2 $\cancel{2}$, USNM), Mount Gay Est. (Leeward side), H.H. Smith (1^Q, USNM), St. Andrew, Mirabeau Est. (Windward side), H.H. Smith (1⁽²⁾, USNM), St. George's (Leeward side), Botanic gardens, 10 September, H.H. Smith (1³, USNM). **GUADELOUPE**: Grande Terre, July 1963, J. Maldonado C. (1³, 1², USNM). **GUATEMALA**: Petén Department, Petén, September 1959, N.L.H. Krauss (2⁽²⁾, USNM), Tikal, September 1959, N.L.H. Krauss (2Å, USNM), Quetzaltenango Department, San Felipe, La Jardin Restaurant and guesthouses, elevation 755 meters, 15 February 2007, A.T. Gonzon (4Å, UDCC). GUYANA: Demerara-Mahaica Region, Demerara River Bank, 1 mile from Georgetown, 22 September 1918, Harold Morrison (1⁽²⁾, USNM), near Peter's Hall, 2 mi. from Georgetown, 22 September 1918, H. Morrison (1⁽²⁾, USNM). HAITI: Portau-Prince, December (2Å, USNM), February (6Å, USNM). HONDURAS: Alta Verapaz near Yiquiche, 16 July 2001, R.L. Snyder (1⁽²⁾, UDCC). JAMAICA: Gordon Th., 1 February 1937, Chapin and Blackwelder (1♂, USNM), Trinity Ville, 28 February 1937, Chapin and Blackwelder (2^{\uparrow}_{\circ} , USNM), Clarendon Parish, Cockpit City, 28 December 1960, J. Maldonado C. (1 \bigcirc , USNM), 28 December 1961 (1 \bigcirc , USNM), Kingston Parish, Blue Mountains, Whitfield Hall, July 1984, N.L.H. Krauss (1 \bigcirc , USNM), Kingston, 9-14 September 1917, Harold Morrison (2 \bigcirc , USNM), St. Ann Parish, Fern Gully, 14 September 1917, Harold Morrison (5♂, 3♀, USNM), Hardwar Gap, 21 July 1968, J. Maldonado C. (1Å, USNM), 6-8 December 1975, Gary

F. Hevel (2Å, USNM), 4 miles north Moneague, 2 February 1937, Chapin and

Blackwelder (4Å, USNM), <u>St. Catherine Parish</u>, Spanish Town, 12 December 1967,

N.L.H. Krauss (1⁽²⁾, USNM), <u>St. Thomas Parish</u>, Morant Point, December 1961, J.

Maldonado C. (1⁽¹⁾, USNM), <u>Trelawney Parish</u>, Clarks Town, 16 February 1937,

Chapin and Blackwelder (1^Q, USNM). **MARTINIQUE**: Lamantin, June 1962, J.

Maldonado C. (4♂, USNM), St. Pierre, Nov. 1950, N.L.H. Krauss (2♂, USNM).

MEXICO: <u>Jalisco</u>, Puerto Vallarta, 5 October 1984, G.E. Bohart (13, 12, 1) broken,

BYUC), Rio Ayuquilla circa Zenzontla, 800 m, 13 October 2001, C.H. Dietrich,

sweeping (2Å, 1^Q, UDCC), <u>Veracruz</u>, 3 miles east Huatusco, 22 July 1995, J.B.

Woolley and G. Zolnerowich (1^Q, TAMU), Cordoba, 22 November 1963, N.L.H.

Krauss (1[♀], USNM). **NICARAGUA**: Musawas, Waspuc River, 23 October 1955, B.

Malkin (2Å, UDCC), Rio San Juan Province, Refugio Bartolo, 51 meters,

10.97254°N, 0.8433906°W, 5-15 August 2002, R.M. Caesar (1∂, TAMU).

PANAMA: 10 November 1952, F.S. Blanton (2∂, USNM), Barro Colorado Island,

N.L.H. Krauss, January 1947 (1³, USNM), Chepo, 25 September 1952, F.S. Blanton

(3♂, 3♀, USNM), Indio-hydrographic station, Canal Zone, N.L.H. Krauss, October

1946 (1⁽²⁾, USNM), Rio las Lajas near Coronado Beach, 17 September 1952, F.S.

Blanton (1⁽²⁾, USNM), <u>Colon Province</u>, Flat rock above Juan Mina, 5 miles up

Chagres River, Canal Zone, 24 August 1918, H. Morrison (1∂, USNM), Mindi Dairy,

Canal Zone, 3 December 1951, F.S. Blanton (9 3° , 2 9° , USNM), Mojinga Swamp,

Canal Zone, 8 November 1951, F.S. Blanton (1[♀], USNM), Darién Province, El Real,

8 August 1952, F.S. Blanton (1⁽²⁾, USNM), <u>Panama Province</u>, Las Cumbres, 26-28

July 1971, M. Daykin (1 \circlearrowleft , 1 \bigcirc , CDAE), light trap, 8 January 1973, H. Wolda (2 \bigcirc ,

USNM), Paja, 13 October 1952, F.S. Blanton (1^d, USNM). PERU: Madre de Dios

Region, Rio Tambopata, Posada Amazonas, S12°48 08.4, W69°17 59.4, September

2004, J.R. Cryan and J.M. Urban (13, North Carolina Museum of Natural Sciences,

New York State Museum Genbank #04-04-02-06). PUERTO RICO: Bayamon,

January 1899, A. Busck (1♂, 1♀, USNM), 13 November 1947 (1♀, USNM), El

Yunque, 20-22 March 1954, J. Maldonado Capriles (1⁽²⁾, USNM), Guajataca Forest,

Isabela, 22 July 1955, collected at light, Ramos and Maldonado (1∂, USNM), Gurabo,

4 November 1944 (1∂, USNM), Luquillo Forest, 30 December 1962, Paul and Phyllis

Spangler (1♂, USNM), Maricao, 2 July 1917, Harold Morrison (2♂, USNM),

Mayaguez, 4 July 1917, Harold Morrison (1 broken, USNM), Mayaguez, 9 October

1935, collected at light (13, USNM), Mayaguez, March 1959, H. Mendoza (13,

USNM), Mayaguez, September-November 1960, M.M. Beauchamp (1^Q, USNM),

Mayaguez, Fed. Exp. Sta. 10 October 1975, E. Freytag (8∂, UDCC), Punta Cangrejos,

22 March 1920, G.N. Wolcott (1², USNM), Yauco-Lares Road, Kilometer 22, 18

July 1953, J.A. Ramos, J. Maldonado, at light (1♂, USNM), Kilometer 29, 20 January

1954 (2♂, USNM). **ST. LUCIA**: Gastries, 10-22 September 1919, J. C. Bradley (14♂,

CUIC). TRINIDAD AND TOBAGO: Aripo savanna, 26 October 1918, Harold

Morrison (13, USNM), Caroni River, 12 October 1918, Harold Morrison (133,

USNM), Single Research Station, sweeping yard, 28 June 1987, T. Myers (23,

UDCC), Port of Spain City Corporation, Botanical Garden, 13 October 1918, Harold

Morrison (13, 14, 1 broken, USNM), D'Abadie, 15 October 1918, Harold Morrison

(6Å, USNM), Department of Agriculture grounds, 24 October 1918, Harold Morrison (7Å, USNM), Savanna, St. Clair, 24 October 1918, Harold Morrison (5Å, 1¢, USNM), <u>San Fernando City Corporation</u>, Golconda estate, 19 October 1918, Harold Morrison (1Å, USNM), Tobago, Archibald Estate, Roxborough, 6 November 1918, Harold Morrison (1Å, USNM), St. George County, Arima, Blanchesuisse road 8th mile, 29 October 1918, Harold Morrison (1¢, USNM). **VENEZUELA**: <u>Amazonas</u>, Aqua Linda, 18-20 June 2000, P. Freytag et al, sweep (1¢, UDCC), T.F.A. Rio Negro, San Carlos de Rio Negro, 5-12 March 1984, O. Flint and J. Louton (1¢, USNM), <u>Lara</u>, Jiménez, Quíbor, 8 July 1979, R.W. Brooks, A.A. Grigarick, J. McLaughlin, and R.O. Schuster (1¢, CDAE), <u>Miranda</u>, Venezuelan Institute for Scientific Research, Altos de Pipe, 2 July 1968, J. Maldonado C. (1Å, USNM), <u>Zulia</u>, Puerto Tarra, Encontrada, January 1970, J. Maldonado C. (1¢, USNM), Maracaibo, Caño Colorado, 27 June 1979, R.W. Brooks, A.A. Grigarick, J. McLaughlin, and R.O. Schuster (1Å,

Flavoclypeus andromedus

USA: ALABAMA: <u>Houston Co</u>., Dothan, Landmark Park, 10 September 2005, L.R. Donovall, sweeping, mowed grasses/sedges (4♂, 4♀, UDCC). ARKANSAS: <u>Logan</u> <u>Co.</u>, Paris, 9 November 1977 ("9-11-77", 1♀, LSAM). DELAWARE: <u>New Castle</u> <u>Co.</u>, Blackbird State Forest, Peters Tract near Saw Mill Road, N39 20 35 W75 44 37, 12 September 2005, A. Gonzon, sweep understory/grass (1♀, UDCC), Newark, Iron Hill Park, 15 July 2004, C.R. Bartlett, sweeping grass & sedges (2♂, UDCC), Iron

Hill Park, 4 August 2004, A. Gonzon, sweeping sedges and grass including *Eleocharis* (1⁽¹⁾, UDCC). DISTRICT OF COLUMBIA: Washington, 14 August 1937, P.W. Oman (2Å, USNM). FLORIDA: Alachua Co., near Gainesville, Paynes Prairie Preserve State Park, near Lake Wauberg, 29.53208, -82.29863, 23 January 2009, sweep grassy vegetation, C.R. Bartlett (11 $^{\circ}$, 6 $^{\circ}$, UDCC); Broward Co., Fort Lauderdale, Hugh T. Birch Recreation Area, sweep, 26 December 1999, C.R. Bartlett (1∂, 3♀, UDCC), Fort Lauderdale, Lauderhill, Invarray, 24 July 1999, C. R. Bartlett (2^Q, UDCC); Fort Lauderdale, Sunrise, NW 20th CT, 26 December 1999, C.R. Bartlett, sweeping grass and weeds near canal (2^{\uparrow} , 4° , UDCC); Fort Lauderdale, H.T. Birch Recreation Area, 26 July 1999, C.R. Bartlett, sweep lawn/weeds (8^Q, UDCC); Highlands Co., near Lake Placid, Archbold Biological Station, 21 January 2002, C.R. Bartlett (13, 29)UDCC) ; Jefferson Co., Wacissa, at Jct SR259 & 60, 27 July 2000, C.R. Bartlett, sweeping roadside (1⁽¹), UDCC); Miami-Dade Co., Airport Fumigation Site, 25 47 58 N 80 18 26 W, 17 October 2008, T. Dobbs, light trap (1♀, UDCC), Miami, 17 October 2003, C. Beal, sweep grass $(1^{\circ}_{+}, UDCC)$; Palm Beach Co., nr. Boca Raton, Loxahatchee Road, 22 January 2002, C.R. Bartlett, sweeping roadside (13, 69), UDCC); near West Palm Beach, Loxahatchee Road, roadside, 22 January 2002, C.R. Bartlett (2^{\uparrow} , 4^{\bigcirc} , UDCC); near West Palm Beach, Seminole Palms Park, 23 January 2002, C.R. Bartlett (1^Q, UDCC), Sarasota Co., Myakka River State Park, 3 September 1954, H.V. Weems (1∂, USNM). LOUISIANA: East Baton Rouge Parish, Baker, Maw Maws house, 14 September 2002, Mindy Pierson, caught by net (1 \bigcirc , UDCC). MARYLAND: Allegany Co., Little Orleans at Little Orleans campground, N39

37.844 W 078 23.348, 5 June 2004, C. Bartlett and A. Gonzon (1⁽²⁾, UDCC); Cecil Co., Fair Hill, Fair Hill Natural Resources Area, 26 September 2003, C.R. Bartlett, sweeping field (18^A, 21^Q, UDCC); same, 24 September 2004, A. Gonzon, sweeping $(1^{\circ}, UDCC)$; same, 18 September 2009, sweeping, C.R. Bartlett (4 $^{\circ}$, 1 $^{\circ}$, UDCC); same, 30 September 2011, sweep, A. Kennedy (6° , 10° , UDCC), Harford Co., circa 2 miles northwest of Havre de Grace, I-95 Park & Ride, N 39 35 804 W 76 08 001, 10 September 2004, A. Gonzon, sweeping grasses (1⁽¹⁾, UDCC). NEW JERSEY: Salem Co., near Salem, 166 Maskell Mill Road, 16 September 2000, C.R. Bartlett, sweeping lawn (6 \cancel{c} , 4 \cancel{c} , UDCC); 21 July 2001, C.R. Bartlett, sweeping (2 \cancel{c} , 1 \cancel{c} , UDCC); 23 August 2003, C.R. Bartlett (1 $\cancel{3}$, 2 $\cancel{2}$, UDCC). NORTH CAROLINA: Brunswick Co., Bald Head Island, 2-4 July 2007, N.H. Nazdrowicz, sweeping (4Å, UDCC), Haywood Co., Great Smoky Mountains National Park, Purchase Knob ATBI house at Appalachian Highlands Science Learning Center, N35 35.222 W83 04.460, 22 June 2006, elevation 1517 meters, C. Bartlett & A. Gonzon, light & night sweep (2^{\bigcirc}) , UDCC), Swain Co., Great Smoky Mountains National Park, Andrew's Bald circa 1.8 miles from Clingman's Dome parking lot, 1707 meters, N35 32.508 W83 29.591, 20 June 2006, C.R. Bartlett and A.T. Gonzon, sweeping grassy bald (1, UDCC), Clingman's Dome Road circa 2.25 miles from US 441, 1706 meters, N35 35.741 W83 27.519, 20 June 2006, C.R. Bartlett and A.T. Gonzon, sweeping roadside grasses (12, UDCC), Wake Co., Raleigh, 16 October 1938, Oman (43° , USNM). PENNSYLVANIA: Chester Co., near Toughkenamon, Stroud Water Research Center, 17 September 2004, A. Gonzon, sweeping (1° , UDCC). TENNESSEE: Blount Co.,

near Townsend, Great Smoky Mountains National Park, Cades Cove at campground, 8 July 2002, C.R. Bartlett et al. (3♀, UDCC); same, Cades Cove, Forge Creek Road, wet meadow, 10 July 2002, C.R. Bartlett et al. (1♀, UDCC); Great Smoky Mountains National Park, Middle Prong, Little River roadside, 10 July 2002, C.R. Bartlett et al. (1♀, UDCC); Great Smoky Mountains National Park, Gregory Bald, 11 July 2002, C.R. Bartlett et al. (2♀, UDCC). TEXAS: <u>Nacogdoches Co.</u>, Nacogdoches, 22 September 1979, M. Klass (1♀, LSAM). VIRGINIA: <u>Accomack Co</u>., Wallops Island, 25 May 1913, W.L. McAtee (1♂, UDCC); <u>Fairfax Co</u>., Vienna, 2 September 1946, P.W. Oman (2♂, ISNB), Vienna, 2 September 1946, P.W. Oman (3♂, USNM).

Flavoclypeus nigrifacies

USA: FLORIDA: <u>Broward Co</u>., Fort Lauderdale, Hugh T. Birch Recreation Area, 26 December 1999 (3♂, 2♀, UDCC), Fort Lauderdale, Lauderhill, Invarray, 24 December 1999, C.R. Bartlett, sweeping lawn/sedge in shallow ditch (3♂, UDCC), same, 5 July 2000 (3♂, UDCC), same, 19 January 2001 (1♂, UDCC), <u>Highlands Co</u>., near Lake Placid, Archbold Biological Station, 21 January 2002, C.R. Bartlett, sweep (17♂, 6♀, UDCC), <u>Jefferson Co</u>., 2 miles south Wacissa, 27 June 2000, C.R. Bartlett (1♀, UDCC). **BELIZE:** <u>Cayo District</u>, near Teakettle Bank, Pook's Hill, 5-6 January 2003, C.R. Bartlett, sweep (5♂, 3♀, UDCC), same, 17 09.257N 88 51.091W, 279 feet, 6 July 2003, C.R. Bartlett (1♀, UDCC), <u>Stann Creek District</u>, just south of Hopkins, 7 January 2003, C.R. Bartlett, shore vegetation (1♀, UDCC). **BOLIVIA**: <u>Santa Cruz</u> <u>Department</u>, 10 miles west Portachuelo, 27 March 1978, UV trap, G.B. Marshall (1♂, LBOB), Est. Exp. Saavedra 250 m, 9 August 1980, D. Foster (13, 19, UDCC).

COLOMBIA: Meta Department, Puerto Lopez, 9 March 1971, S.S. Roback (1^Q, USNM). COSTA RICA: Cartago Province, Pejibaye, 24-25 March 1987, W.E. Steiner, yellow pan trap in old field and agricultural area (1° , USNM), Guanacasta Province, Estación Experimental Enrique, Jiménez Munez, January 1993, F. Parker (1Å, LBOB), Heredia Province, 10 August 1975, N.L.H. Krauss (1Å, USNM), near Puerto Viejo La Selva Biological Station 179 ft N10°25 W84°00, C.R. Bartlett, J. Cryan, J. Urban, 15-17 August 2003 (123, 202, UDCC), C.R. Bartlett et al, 24 February 2004 (12³, 13², UDCC), Limón Province, 24 kilometers southeast Limón, at light, 4 August 1990, W.F. Chamberlain (2^Q, TAMU), Puntarenas Province, Brujo, 7 August 1990, G.M. Chamberlain (1^A, TAMU). ECUADOR: Orellana Province, Yasuni National Park, S00°40.478 W76°23.866, 26-29 May 2005, C.R. Bartlett, N. Nazdrowicz, D. Chang, sweeping/day (13Å, 11°, UDCC). GUYANA: Demerara-Mahaica, near Peter's Hall 2 miles from Georgetown, 22 September 1918, H. Morrison (13, USNM). **JAMAICA**: Kensworth, 18 February 1937, Chapin and Blackwelder (1^Q, USNM), <u>Trelawney Parish</u>, Clarks Town, 16 February 1937, Chapin and Blackwelder (23, 12, USNM). **MARTINIQUE**: Fort-de-France, November 1950, N.L.H. Krauss (1³, USNM), Saint-Pierre, November 1950, N.L.H. Krauss (1⁽²⁾, USNM). **MEXICO**: Federal District, Mexico City area, 1940's, D.M. DeLong (6 $^{\circ}$, 3 $^{\circ}$, UKYC), Veracruz, 3 miles east Huatusco, 22 July 1995, J.B. Woolley and G. Zolnerowich (5 $\cancel{3}$, 3 $\cancel{2}$, TAMU). **PANAMA**: Chiriquí Province, David, N.L.H. Krauss, December 1946 (1° , USNM), Gualaca, 14 December 1952,

F.S. Blanton (1^Q, USNM), <u>Panama Province</u>, Tocumen, 4 February 1953, F.S.

Blanton (1⁽¹), USNM), Veraguas, Cerro Tute, 4 kilometers west Santa Fe, 680 meters,

2 August 1995, C.W. and L. O'Brien (1^Q, LBOB). **PUERTO RICO**: Isabela,

Guajataca Forest, 22 July 1955, collected at light, Ramos and Maldonado (13,

USNM), Mayaguez, Federal Experiment Station, 10 October 1975, P.F. Freytag (23³),

18♀, UKYC). **ST. THOMAS**: 27-30 March 1961, J. Maldonado C. (1♂, USNM). **ST.**

VINCENT: H.H. Smith, 18, P.R. Uhler collection (13, 19, USNM). **VENEZUELA**:

Amazonas, Agua Linda River, 5°49'5"N 67°27'29"W, 18-20 June 2000, sweep, P.

Freytag, M.A. Gaiani, Q. Arias (2⁽²⁾, 3⁽²⁾ UDCC), <u>Apure</u>, near San Fernando de Apure,

7 50'44"N 67 29' 10"W, 20 June 2000, blacklight, P.M. Freytag, M.A. Gaiani, and Q.

Arias (1 $\stackrel{\bigcirc}{+}$, UKYC), same (10 $\stackrel{\bigcirc}{-}$, 16 $\stackrel{\bigcirc}{+}$, UDCC), <u>Aragua</u>, Rancho Grande, Henry Pittier

National Park, 1100 meters, 24 December 1985, P. Kovarik, R. Jones (3^Q, TAMU),

Barinas, 5 kilometers east Altamira de Caceras, 700 meters, 30 December 1985, P.

Kovarick, R. Jones (1^o, TAMU), <u>Guarico</u>, 5 kilometers north Santa Rita, 400'. 28

July 1989, C. & L. O'Brien and G.J. Wibmer (1⁽²⁾, LBOB), 7 kilometers east-southeast

Calabozo, Est. Biol. Llanos, 380', 21 July 1988, C.W. and L. O'Brien and G.J.

Wibmer (1♂, LBOB), north Calabozo Dam, 350', 22 July 1988, C. and L. O'Brien and G. Wibmer (1♂, LBOB).

Flavoclypeus nitens

USA: DELAWARE: <u>New Castle Co.</u>, Ashland near Ashland Nature Center along Red Clay Creek, 1 September 2005, A.T. Gonzon at mercury lamp (2♂, UDCC), same,

C.R. Bartlett at mercury lamp (1 $^{\circ}$, 2 $^{\circ}$, UDCC), Newark, White Clay Creek Preserve, August 2005, at light (1∂, 2♀, UDCC). DISTRICT OF COLUMBIA, Washington, 15 May 1931, P.W. Oman (1⁽²⁾, USNM), 15 April 1934, P.W. Oman (1 broken, USNM). ILLINOIS: Coles Co., Charleston, 3 May 1943 (1⁽²⁾, USNM), Piatt Co., Sangamon River, 7 miles northeast Monticello, 16 May 1936 (2⁽²⁾, USNM). MARYLAND: Allegany Co., Little Orleans, Little Orleans campground, N39 37.83 W78 23.36, 6-7 June 2008, C.R. Bartlett (1 $^{\circ}$, UDCC), same, sweep/aspirator, damaged (1 $^{\circ}$, UDCC), Montgomery Co., near Chevy Chase Lake, 6 July 1913, W.L. McAtee (1^Q, USNM), Plummers Island, 18 May 1913, R.C. Shannon (1^Q, USNM), 13 July 1913, R.C. Shannon (1 \bigcirc , USNM), 28 April 1914, R.C. Shannon (1 \bigcirc , USNM). NORTH CAROLINA: Haywood Co., Great Smoky Mountains National Park, circa 0.8 mile SSE of Purchase Knob ATBI house, along gravel drive, 1417 meters, N35 34.889 W83 04.214, 22 June 2006, C.R. Bartlett and A.T. Gonzon, sweeping grassy meadow & bank (1⁽²⁾, UDCC), Great Smoky Mountains National Park, circa 1 mile SSE of Purchase Knob, ATBI house along gravel drive, 1381 meters, N35 34.736 W83 04.132, 22 June 2006, C.R. Bartlett and A.T. Gonzon, sweep grassy meadow and roadside (53, 12, UDCC). TENNESSEE: Blount Co., Great Smoky Mountains National Park, Cades Cove near Abrams Creek, elevation 526 meters, circa 0.3 miles from CC Loop Road, N35 35.367 W83 50.274, 21 June 2006, C.R. Bartlett and A.T. Gonzon, sweeping grass sedge in wet meadow ($3^{\uparrow}_{\circ}, 3^{\bigcirc}_{+}, \text{UDCC}$), near Townsend, Great Smoky Mountains National Park, Cades Cove, Forge Creek Road, Wet Meadow, 8 July 2002, C.R. Bartlett et al. (23° , UDCC), near Townsend, Great Smoky

Mountains National Park, Cades Cove, Laurel Creek Road, 10 July 2002, C.R. Bartlett (1^Q, UDCC). TEXAS: <u>Gonzalez Co.</u>, Palmetto State Park, 1 June 1984, J.B. Woolley (5^A, TAMU), <u>San Patricio Co.</u>, 15 kilometers northeast Sinton, Welder Refuge, 28°06.9'N, 97°23.9'W, 5 meters, 1-8 April 2004, S. & J. Peck, riparian woodland, lot 17 (1^A, TAMU). **MEXICO**: <u>Coahuila</u>, 7 miles south-southwest Saltillo, 4 July 1984, J.B. Woolley (2^A, TAMU).

APPENDIX B

QIAGEN DNEASY DNA EXTRACTION PROTOCOL

Set the water bath to 56°C.

Indicate on the NYSM Genome Bank master list which specimens you plan on using and the appropriate specimen numbers (ex. DEL175).

Label 3 1.5 ml microcentrifuge tubes for each specimen.

Label a kim wipe with the specimen numbers and place specimens on kim wipe to blot dry.

Remove one hind leg of delphacid with scalpel and place in labeled microcentrifuge tube. Wipe pin with ethanol between each cut.

Add 180 µl of ATL buffer to each sample.

Add 20 µl of Proteinase K to each sample, shake bottle before adding.

Vortex samples briefly and place in water bath for 6+ hours.

Remove specimen from sample and place back in NYSM GB vial to retain as voucher (wipe forceps with ethanol each time.)

Vortex.

Add 200 µl AL buffer to each sample and vortex.

Add 200 µl 100% EtOH to each sample and vortex.

Set up and label spin columns for each sample.

Pipet entire contents of sample into spin column with 2 ml collection tube (set pipet to approximately $610 \ \mu l$ for this).

Centrifuge at 8,000 rpm for 1 min.

Discard flow through and place spin column in a new collection tube.

Add 500 µl of AW1 buffer and centrifuge at 8,000 rpm for 1 min.

Discard flow through and place spin column in a new collection tube.

Add 500 µl of AW2 buffer and centrifuge at 14,000 rpm for 3 min.

Discard flow through and place spin column in a labeled 1.5 ml centrifuge tube.

Add 200 µl of AE buffer, wait one minute, then centrifuge at 8,000 rpm for 1 min. Remove spin column from microcentrifuge tube, do not discard elution, and place in

new microcentrifuge tube.

Add 200 μ l of AE buffer, wait one minute, then centrifuge at 8,000 rpm for 1 min. Combine both elutions and pipet ~40 μ l into a separate labeled tube to use for PCR. Store at -80°C.

APPENDIX C

COI PCR PROTOCOL

Make enough master mix for the number of reactions you wish to do. *Make sure you add the Taq last!

Master Mix for 1 Rxn	
10x PCR Buffer	2.5µl
25mM MgCl ₂	2.5µl
10mM dNTPs	2.0µl
Primer 1	0.5µl
Primer 2	0.5µl
Taq Polymerase	0.2µl
Pure H ₂ O	16.5µl

Finger vortex the tube and place on ice.

If you have not done so already, record where each sample will be on the PCR plate/strip.

Label your PCR plate/strip.

Add 23μ l of Master Mix to each PCR tube. Add 25μ l of Master Mix to your control Add 2μ l of genomic template to each tube.

Put the caps on and make sure the liquid is not clinging to the sides.

Place in the thermocycler and run the Ron Calvin protocol.

Ron Calvin protocol:

Temperature Control mode: calculated

Lid control mode: tracking at 4°C

Cycle 1:	94°C for 3 minutes	x1
Cycle 2:	94°C for 1 minute	
	45°C for 1 minute	
	72°C for 1 minute	x35
Cycle 3:	72°C for 10 minutes	x1
Cycle 4:	4°C (holding)	

APPENDIX D

ELECTROPHORESIS PROTOCOL

Prepping gel: (small/medium/large)

Measure 50/150/450 ml of 0.5x TBE solution and pour into bottle or beaker.

Weigh ~0.75/2.25/6.75 g of agarose and add to TBE.

Microwave with lid on loosely. Watch and frequently swirl to ensure mixture does not boil over.

When agarose is fully dissolved let it stand for a 2-3 mins.

Add 7.5/20/60 µl of EtBr to agarose and mix well.

Set up tray so agarose does not leak out and with desired wells.

Allow agarose to continue cooling until the bottle can be handled.

Pour agarose into tray slowly from the corner and allow to set (approximately 15 mins).

Rotate tray and cover gel with 0.5x TBE solution.

Loading gel: (small wells/large wells)

Cover empty tips tray with parafilm and run fingers over to create indentations, each depression used will be a well on the gel.

Place $1/2 \ \mu$ l of loading dye in each depression except for the depressions that will carry ladders.

Place $6/12 \mu l$ of ladder in the appropriate depressions.

Add $5/10 \ \mu$ l of the appropriate genomic template to each depression; use a new tip each time.

Load the gel rinsing pipet between lanes.

Run gel:

Run at 100 v for approximately 1 hour.

APPENDIX E

ETHANOL PRECIPITATION PROTOCOL

Bring PCR volume up to 100 microliters with dH₂O Add 10% 5M NaCl Add 3 volumes 100% EtOH Freeze at -80° for 30 minutes Store at -20° overnight Spin 30 minutes at top speed Carefully pour off supernatant; ensure pellet is not dislodged (pellet will be small and white, just visible) Add 500 microliters cold (-20°) 70% EtOH Spin 5 minutes at top speed Carefully pour off supernatant; ensure the pellet is not dislodged Let dry (~2 hours) Resuspend with 20-30 microliters ddH₂O/nuclease-free water; pipet the sample up and down several times to ensure the pellet goes into solution.

APPENDIX F

TEXT OF NEXUS FILE FOR PHYLOGENETIC ANALYSIS

#NEXUS
[execute C:\Users\Ashley\Desktop\coded_chars.nex;]

begin taxa; dimensions ntax=18; taxlabels c_atridorsum c_nigriscutellatus c_teapae ch_havanae d adunca d_andromeda d aterrima d_balli d_incurva d_latidens d_livida d_nigrifacies d nitens d_recurvata d_shermani d sucinea k lutulenta m_arvensis end; begin characters; dimensions nchar=34; format missing=? symbols="01234" equate="-=?"; matrix c atridorsum 10011 10101 00101 31100 01011 01110 0010 100?3 10111 00001 10210 11111 11110 0010 c_nigriscutellatus 03110 12210 12112 01003 12110 01000 0110 c_teapae ch_havanae 011?0 10001 13201 00103 30110 00--0 0001 12001 11211 01021 10310 02211 01011 1010 d adunca 03001 01111 02030 30111 02211 01101 1010 d andromeda d aterrima 13120 13211 02223 11200 ?001? 10--0 0000

d_balli d_incurva d_latidens d_livida d_nigrifacies d_nitens d_recurvata d_shermani d_sucinea k_lutulenta m_arvensis ; end;	22000 11122 03110 02001	12211 12111 12111 01111 11111 11111 12210 13101 10001	01122 01010 13011 02001 02013 13023 03030 02021 11001	20210 10212 11001 00112 11201 11310 11312 21110 01201	<pre>?120? 01101 ?110? 12001 11211 00100 ?201? ?0000 0020?</pre>	01110 001 11010 000 -00	0011 0010 0010 0010 0110 0001
begin assumptions;							
typeset types	= Ord: 1	2345	57812	14 15 1	6 18 20) 21;	
; end;							
begin paup; assume types	et=tvnes•						
set maxtrees=	• -		uto noti	fybeep=	=no erro	orbeep=1	no;
log start file=	•	-	4				
outgroup ch_havanae k_lutulenta m_arvensis; cstatus full=yes;							
showmatrix;							
hsearch adds showtrees;	eq=rando	m nrep	s=1000	;			
bootstrap nre					lseq=rai	ndom ni	reps=10;
hsearch adds	eq=rando	m nrep	s=1000	;			
reweight; hsearch addseq=random nreps=1000;							
reweight;							
hsearch addseq=random nreps=1000; reweight;							
hsearch addseq=random nreps=1000;							
reweight; hsearch addseq=random nreps=1000;							
showtrees;							
describetrees /plot=phylogram apolist=yes; cstatus;							
bootstrap nreps=1000 wts=proportional /addseq=random nreps=20; savetrees file=tree.tre brlens=yes savebootp=both from=1 to=1;							
savetrees file	=tree.tre	brlens=	yes sav	ebootp	=both fi	rom=1 t	o=1;

log stop;

end;