EFFECTS OF HIGH-FREQUENCY, LOW-MAGNITUDE VIBRATION ON MUSCLE ACTIVITY IN CHILDREN WITH SPASTIC CEREBRAL PALSY

by

Victoria L. Haggett

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

Spring 2015

© 2015 Victoria L. Haggett
All Rights Reserved
EFFECTS OF HIGH-FREQUENCY, LOW-MAGNITUDE VIBRATION ON MUSCLE ACTIVITY IN CHILDREN WITH SPASTIC CEREBRAL PALSY

by

Victoria L. Haggett

Approved:

Christopher M. Modlesky, Ph.D.
Professor in charge of thesis on behalf of the Advisory Committee

Approved:

William Farquhar, Ph.D.
Chair of the Department of Kinesiology and Applied Physiology

Approved:

Kathleen Matt, Ph.D.
Dean of the College of Health Sciences

Approved:

James G. Richards, Ph.D.
Vice Provost for Graduate and Professional Education
ACKNOWLEDGMENTS

Christopher M. Modlesky, Ph.D., for advising me through my thesis and providing opportunities to progress my knowledge and skills in the field of kinesiology and applied physiology.

Christopher Knight, Ph.D., for his contribution to the data collection and processing aspects of my thesis, teaching me the skills related to these parts of my thesis, for providing valuable input on my thesis, and for being on my thesis committee.

Kurt Manal, Ph.D., for his support and input regarding data collection for my thesis and for being on my thesis committee.

Freeman Miller, MD, for his support and input regarding the patient population for my thesis and for being on my thesis committee.

Harshvardhan Singh, M.S., BPT, for providing guidance throughout all aspects of my thesis and master’s education.
Daniel Whitney, M.S., for his help through data collection and writing my thesis and providing guidance throughout my master’s education.

My parents and grandparents, for the support they have given me throughout my education.

The Eunice Kennedy Shriver National Institute of Child Health and Human Development, NIH (Grant R15HD071397-01)
# TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>LIST OF TABLES</td>
<td>vii</td>
</tr>
<tr>
<td>LIST OF FIGURES</td>
<td>viii</td>
</tr>
<tr>
<td>ABSTRACT</td>
<td>ix</td>
</tr>
<tr>
<td>Chapter</td>
<td></td>
</tr>
<tr>
<td>1 INTRODUCTION</td>
<td></td>
</tr>
<tr>
<td>Overview</td>
<td>1</td>
</tr>
<tr>
<td>Significance</td>
<td>2</td>
</tr>
<tr>
<td>Purpose</td>
<td>2</td>
</tr>
<tr>
<td>Hypotheses</td>
<td>2</td>
</tr>
<tr>
<td>Limitations</td>
<td>2</td>
</tr>
<tr>
<td>2 LITERATURE REVIEW</td>
<td>4</td>
</tr>
<tr>
<td>Overview of Complications Associated with Cerebral Palsy</td>
<td>4</td>
</tr>
<tr>
<td>Applications for Vibration</td>
<td>5</td>
</tr>
<tr>
<td>Mechanisms of Vibration</td>
<td>9</td>
</tr>
<tr>
<td>Electromyography Methodology with Vibration</td>
<td>14</td>
</tr>
<tr>
<td>3 MANUSCRIPT</td>
<td>16</td>
</tr>
<tr>
<td>Introduction</td>
<td>16</td>
</tr>
<tr>
<td>Methods</td>
<td>18</td>
</tr>
<tr>
<td>Participants</td>
<td>18</td>
</tr>
<tr>
<td>Anthropometrics</td>
<td>18</td>
</tr>
<tr>
<td>Tanner Staging</td>
<td>19</td>
</tr>
<tr>
<td>Gross Motor Function Classification System (GMFCS)</td>
<td>19</td>
</tr>
<tr>
<td>Modified Ashworth Scale (MAS)</td>
<td>19</td>
</tr>
<tr>
<td>Instrumentation</td>
<td>19</td>
</tr>
<tr>
<td>Data Collection</td>
<td>20</td>
</tr>
<tr>
<td>Data Analysis</td>
<td>23</td>
</tr>
<tr>
<td>Statistical Analysis</td>
<td>24</td>
</tr>
<tr>
<td>Results</td>
<td>24</td>
</tr>
</tbody>
</table>
Discussion..................................................................................................................... 29
Conclusions .................................................................................................................. 33

REFERENCES ............................................................................................................. 35

Appendix

A  GROSS MOTOR FUNCTION CLASSIFICATION SCALE ......................... 41
B  INSTITUTIONAL REVIEW BOARD APPROVAL LETTERS ..................... 42
C  FREQUENCY FILTERING TECHNIQUE......................................................... 45
LIST OF TABLES

Table 1  Subject descriptives for both CP and TD groups are presented, including anthropometrics, Tanner stage, Gross Motor Function Classification System (GMFCS), Modified Ashworth Scale grade (MAS), and maximal voluntary isometric contraction torque (MVIC). MVIC was the only variable significantly different between groups. * $p < 0.05$. .................. 26
LIST OF FIGURES

Figure 1  Anterior (A), posterior (B), and left lateral (C) views of the set-up of a child standing on the HLV platform used in this study. Surface electrodes were affixed to the skin with self-adhesive wrap and tape when able................................................................. 22

Figure 2  Differences between the HLV off condition prior to HLV (PreHLVoff) and the HLV off condition after HLV (PostHLVoff) are presented for each muscle for A: children with CP and B: TD children. There were no significant differences between the two conditions for any muscle in either group................................................................. 27

Figure 3  Mean differences between HLVon and HLVoff are presented for the tibialis anterior (TA), lateral gastrocnemius (LG), medial gastrocnemius (MG), soleus (SOL), biceps femoris (BF), and vastus lateralis (VL). *Condition effect with HLVon > HLVoff for the cerebral palsy (CP) and typically developing (TD) groups combined, p < 0.05. **Group by condition effect with HLVon > HLVoff in the CP group but not in the TD group, p < 0.05................................................................. 28

Figure 4  Frequency spectrum from the LG of a vibration trial where the unfiltered data is represented in black and the filtered data is overlaid represented in grey. ................................................................. 45

Figure 5  EMG data from the LG of a vibration trial where the unfiltered data is represented in black and the filtered data is overlaid represented in grey. ................................................................. 46
ABSTRACT

Cerebral palsy (CP) is one of the most common causes of disability in children and is a neuromuscular disorder that limits mobility, impairs motor control, and is damaging to the musculoskeletal system. One treatment that has shown promise in offsetting the limited musculoskeletal development in children with CP is high-frequency, low-magnitude vibration (HLV). The purpose of this study was to determine if an HLV stimulus emitted by a floor-based platform increases muscle activity in the lower extremities of children with spastic CP. We hypothesized that an HLV stimulus would increase muscle activity in children with spastic CP.

Children with spastic CP between the ages of 4-11 with a gross motor function classification of I-II (n = 11) and TD children of similar age (n=10) took part in this study. Muscle activity was measured for the tibialis anterior (TA), lateral gastrocnemius (LG), medial gastrocnemius (MG), soleus (SOL), biceps femoris (BF), and vastus lateralis (VL) using surface electromyography (EMG). Children stood on a platform that emits an HLV (30 Hz at 0.3 g) for three conditions that lasted thirty seconds each. First, the platform was off (pre-HLV), then on (HLV), then off again (post-HLV). Root mean square (RMS) values were obtained from the EMG data collected from all muscles, and differences between conditions and groups were tested.
There were no significant differences between pre-HLV and post-HLV conditions in children with CP or TD children \((p > 0.05)\). A significant condition effect suggests that there was greater muscle activity during the HLVon condition versus the HLVoff condition in the MG, SOL and BF muscles \((p < 0.05)\). Furthermore, there was a significant group-by-condition effect with greater muscle activity during the HLV condition in the TA, LG, and VL muscles \((p < 0.05)\) for the CP group but not the TD group. The results from this study indicate that a floor-based HLV stimulus increases the activity of the lower extremity muscles in children with spastic CP. The enhanced sensitivity of muscle to HLV in children with spastic CP may explain, at least in part, the previously shown positive effect of HLV on the musculoskeletal system in children with CP.
Chapter 1

INTRODUCTION

Overview

Cerebral palsy (CP) is one of the most common causes of disability in children and is a neuromuscular disorder that limits mobility, impairs motor control, and is damaging to the musculoskeletal system [1, 2]. In children with CP, lower mechanical loading on the skeleton due to poor muscle strength combined with limited mobility results in deficits in structural integrity and bone mass accretion [2, 4]. One treatment that has shown promise in offsetting the limited musculoskeletal development in children with CP is high-frequency, low-magnitude vibration (HLV) [2, 3].

Studies in both animals [5] and humans [6, 7, 8] have reported beneficial osteogenic effects associated with HLV treatment. However, the mechanism by which HLV acts on the musculoskeletal system is still relatively unknown. High-frequency, low-magnitude vibration could act directly on bone cells via reactionary forces produced by the skeleton. Another plausible mechanism is that bone formation is modulated by HLV-induced increases in muscle activity [9]. This HLV-induced muscle activity would likely be due to a spinal reflex where HLV elicits an excitatory response from the muscle spindle, activating motoneurons to resist a falsely detected stretch [10]. Since children with spastic CP already have hyper-excitatory motoneurons [11], it is plausible that this population would have a greater muscle response to HLV than typically developing peers.
Significance

Since CP is one of the most common causes of disability in children [1] and a complication of particular concern is the deterioration of the musculoskeletal system [2, 4], it is critical to identify treatments that counteract the limited muscle and bone development in children with CP. This study is significant because in other populations with compromised musculoskeletal health, HLV has been shown to improve parameters of skeletal integrity [2, 3, 6, 7, 8, 12]. Therefore, it is important to understand the effect of HLV on the musculoskeletal system in children with CP as well as the mechanism behind improvements seen in the musculoskeletal system due to HLV. This is important when considering HLV as a potential non-pharmacological treatment to improve musculoskeletal health in children with CP.

Purpose

The purpose of this study was to determine if an HLV stimulus emitted by a floor-based platform during standing increases muscle activity in the lower extremities in children with spastic CP and if that increase is greater than in typically developing children.

Hypotheses

Hypothesis: An HLV stimulus will increase muscle activity in the lower extremities during standing in children with spastic CP.

Limitations

One limitation to this study is posture. Children with spastic CP often stand differently from each other as well as from typically developing children, largely due to varying degrees of muscle tightness associated with spasticity. Therefore, there is
the limitation of the inability to fully control for posture in this study. However, children with mild to moderate CP who could stand independently participated in this study, so differences in posture were minimized.

Another limitation of this study is generalizability. Since only children with mild to moderate CP participated in this study, findings cannot be generalized to more severe levels of CP. Furthermore a specific floor-based HLV of 30 Hz at 0.3 g was used in this study, so findings cannot be generalized to other types of vibration. However, this specific level of vibration was used because it was determined to be safe as well as effective in showing improvements to the musculoskeletal system in previous studies [2].
Chapter 2
LITERATURE REVIEW

Overview of Complications Associated with Cerebral Palsy

Cerebral palsy (CP) is one of the most common causes of physical disability in children with lifetime costs associated with the disability estimated at 11.5 billion dollars [1]. CP is defined as a group of permanent disorders affecting movement and posture and associated with injury to motor areas of the brain before or shortly after birth [13, 14]. It is a disorder characterized by a variety of motor control problems with severity ranging on a continuum from little motor impairments with full mobility to complete immobilization [1, 14]. The severity of CP is identified using the Gross Motor Classification System, which has five levels and is described in detail in Appendix A [14, 15].

Due to the motor impairments and physical disability associated with CP, children with this disorder tend to have poor muscle strength and control, which limits their daily function [2]. This reduces overall physical activity, and it has been reported that children with CP not only get significantly less physical activity than their typically developing peers but this population also gets less physical activity than recommendation guidelines [16]. This reduction in habitual physical activity and function leads to secondary health issues in this population, such as metabolic dysfunction, cardiovascular disease [16], and inadequate bone structure properties [2, 17].
An important complication in children who have CP is the negative effects on the musculoskeletal system, including low bone mass [18] and underdeveloped bone architecture [18, 19, 20], and a high rate of fracture, especially in the lower extremities [21], further reducing activity. This is most likely due to decreased mobility associated with CP and consequently, decreased mechanical loading on the bones [2, 4]. This results in disuse osteoporosis that further affects quality of life through childhood and adulthood [22]. When a healthy peak bone mass is not reached during growth in childhood when the skeleton is most sensitive to mechanical loading, then there are negative effects on bone quality and strength throughout life [23].

Another complication associated with some types of CP is spasticity, resulting in rigidity of the spastic limb. In the lower extremities, spasticity is characterized as flexor and extensor excitation dependent on stretch velocity and flexor excitation and extensor inhibition dependent on muscle lengthening during stretch [24]. Children with spastic CP are often exposed to treatments, such as physical therapy to improve independent mobility, botulinum toxin injections to relax spastic muscles, and surgery to lengthen tendons and muscles. However, there is no one cure for CP, and a variety of experimental treatments are used to improve mobility and function in children with this disorder [25, 26].

Applications for Vibration

Vibration has been found to be beneficial when used as a modality for treatment and training for a variety of clinical and performance related applications [2, 7, 27, 28]. For children with CP, there are promising findings when using vibration treatment to improve deficits in the musculoskeletal system [2, 3, 4, 6]. Furthermore, findings in stroke patients with spasticity suggest an exaggerated response to vibration
due to a sensitive reflex response [10, 29], which may also be present in children with spastic CP.

In athletes, vibration has been shown to improve performance [27, 28, 30]. Improvements in jump height have been seen in those who have received vibration training [28, 30]. Torvinen et al. [30] found a 7.8% net benefit in vertical jump height in healthy adults who received vibration treatment over a period of eight months. Another study by Bosco et al. [28] found that vibration during training resulted in greater muscle activity. This study done with elite boxers studied electromyogram activity of the arm flexors with and without vibration applied during a loaded bicep curl. When this was done, there was greater muscle activity in the vibrated arms that was more than twice that of the baseline loaded bicep curl measure. This suggests that improvements seen in mechanical power with vibration training in elite athletes may be due to a neurological adaptation. Another study by Cardinale and Lim [31] in elite volleyball players found greater muscle activity in the vastus lateralis due to vibration when performing a half squat. Specifically, a 30 Hz vibration was shown to elicit the highest muscle response, suggesting that this is a frequency that may also elicit the highest reflex response in the vastus lateralis muscle.

Studies in animals have shown promise for vibration as a treatment to strengthen the musculoskeletal system, specifically high-frequency, low-magnitude vibration (HLV) [32, 33, 34]. Rubin et al. [32] examined bone properties in sheep before and after a treatment regimen of floor-based HLV that was given at 0.3 g at 30 Hz and found that the anabolic effect was specific to trabecular bone. In this study, sheep given the HLV treatment for one year had significantly greater bone density (+34.2%) compared to control sheep who did not receive HLV treatment, as well as
greater trabecular bone volume (+32%), greater trabecular mesh number (+45%), and reduced mesh spacing (-36%). Another study by Wolf et al. [33] examined HLV treatment on bone healing in sheep. After an osteotomy in the sheep, an HLV stimulus was applied over a period of eight weeks in a treatment group. Where no significant differences were found between the treatment and control group, callus formation was slightly enhanced in the group that received HLV treatment, and a longer time period of treatment than examined in this study could potentially show differences between groups. Shi et al. [34] studied HLV treatment (35 Hz at 0.3 g) on fracture healing in ovariectomy-induced (OVX) osteoporotic rats over two, four, and eight weeks. Results showed HLV treatment to be effective in promoting fracture healing in the OVX group with greater callus formation, mineralization, and remodeling than the non-OVX group. These studies support HLV as a means to improve bone healing [32, 33, 34], but show that improvements seen in bone due to HLV may be more apparent in diseased bones than in healthy bones.

Studies in humans further support HLV as a treatment to improve musculoskeletal health in osteoporotic and osteopenic populations [4, 6, 7, 8, 12]. Gilsanz et al. [12] studied HLV treatment (30 Hz at 0.3 g) in young women with low bone mineral density (BMD) and found increased bone and muscle mass after one year of regular HLV treatment. In another study by Lam et al. [8], HLV treatment was given for one year in a population of osteopenic girls with adolescent idiopathic scoliosis with results showing HLV-induced improvements in bone in the femur and lumbar spine. HLV treatment (30 Hz at 0.2 g) has also been shown to inhibit bone loss in postmenopausal women, with increasing significance with higher compliance and lower body mass. This suggests that HLV treatment not only improves bone
properties, but can also offset detrimental effects of diseases on the musculoskeletal system [6]. A study by Asselin et al. [4] further supports HLV as a preventative treatment in spinal cord injured (SCI) populations with the finding that the SCI population transmitted vibration signals just as effectively as a healthy population. A review by Rubin, Judex, and Qin [7] supports HLV as an anabolic stimulus to bone that can be used as a passive exercise to improve bone quantity and quality. This review focused on floor-based HLV below 0.5 g, and makes an important note that HLV is safe to be given to humans at 30 Hz at 0.3 g for up to four hours per day, which is 12-24 times that which has shown improvements in bone. This suggests that HLV treatment is a vibration at a low enough level to be safe, yet has potential to be a non-pharmacological avenue for prevention and treatment of osteoporosis.

Specifically, HLV as a non-pharmacological treatment for children with disabilities, including CP, has shown to be effective in offsetting the detrimental effects that disabilities can have on the musculoskeletal system in children [2, 3, 22, 32]. A study by Xie et al. [36] suggested that as little as 10 minutes of floor-based HLV exposure a day could inhibit trabecular bone resorption in the growing skeleton. Katusic and Mejaski-Bosnjak [35] suggested that vibration stimulates the musculoskeletal system giving the need to modulate muscle activity and found significant improvements in motor performance, better stability, and selectivity of movements after vibration treatment on a bed pad. Reyes et al. [22] also found HLV to be a safe and effective treatment to improve bone mass and muscle strength in children with motor disabilities. Another study on children with motor disabilities by Ward et al. [3] found benefits to bone due to HLV treatment. After six months of floor-based HLV treatment (90 Hz at 0.3 g), children who received treatment had a
significantly greater change in volumetric tibial bone mineral density even with a low compliance of 44%. Wren et al. [2] confirmed that six months of HLV treatment (30 Hz at 0.3 g) was effective in improving bone strength in children with CP specifically. This study also used a floor-based HLV and found significant increases in cortical bone properties and bone strength due to HLV treatment. Even though vibration in an occupational setting can be damaging to humans [37], studies have shown and confirmed that floor-based HLV treatment is not harmful to humans and can be beneficial to humans with a damaged musculoskeletal system [6, 30, 38].

**Mechanisms of Vibration**

Where it has clearly been shown that HLV treatment can have beneficial effects on the musculoskeletal system, it is important to understand the underlying mechanisms of how vibration acts on the musculoskeletal system, especially when examining patients with neurological disorders. Some proposed neural mechanisms include a spinal reflex elicited by vibration that enhances muscle contraction, storage and release of mechanical energy by tendons where stiffness can effect transmission, an excitatory response from the muscle spindle detecting changes in length, and a muscle tuning response to reduce vibrations transmitting through soft tissues in the body [10]. Rittweger [27] notes that vibration is a mechanical oscillation that accelerates the human body and may cause a reactive force by and within the body. This review also supports muscle tuning and muscle spindle response as potential mechanisms of how vibration acts on the body.

A study by Pollock et al. [39] looked at single motor units (MU) in the vastus lateralis of healthy adults before and after one-minute bouts of HLV. Results showed a relationship between MU activity and phase of the vibration cycle that suggested that
there is a phase lock of MU firing with the vibration cycle, which indicates that muscle activity seen during vibration could be reflexive. Presynaptic inhibition was found to be unaffected by vibration suggesting that synaptic effectiveness is unlikely to be a cause of changes in muscle activity due to vibration. The authors of this study suggest that vibration causes the primary endings of muscle spindles to respond 1:1, phase locked with the vibration frequency. This data supports muscle spindle response to vibration as a potential mechanism for greater muscle activity seen due to vibration. This would occur when the muscle spindle detects a false change in length due to the vibration and has an excitatory response to resist the stretch, activating muscles [10].

Zaidell et al. [40] looked at an HLV level of vibration to examine a vibration reflex termed the “Tonic Vibration Reflex” (TVR) in a loaded and unloaded leg. Healthy adults were exposed to vibration either floor-based or with Achilles tendon vibration (ATV) with or without an external load, and results indicated that this type of vibration was able to elicit a TVR response in the lower limb muscles when delivered during relaxed sitting or loading. This suggests a spinal reflex, which again could occur through an excitatory response of the muscle spindle, activating Ia afferents that excites alpha motoneurons, which could cause muscle activity either through MU synchronization or inter-muscular coordination where agonists and antagonists are uncoupled [10, 27].

An unpublished study by Suresh et al. [29] examined vibration in a hemispastic stroke population to better understand the role vibration would play in a population with neurological impairments, specifically spasticity. The authors noted that spasticity is characterized by an exaggerated reflex response to muscle stretch or tendon tap, which could make these muscles highly excitable when exposed to
vibration. Therefore, a potential vibration reflex could be useful in assessing the degree of excitability of muscles in patients with spasticity. Results showed that the affected side of the stroke patients had a larger slope, quicker rise to peak force, and greater peak force than the unaffected side. There was a greater response on the affected side in these individuals, suggesting that spasticity may play a role in enhancing muscle response to vibration.

The idea that spasticity induces greater muscle response to vibration is supported by a study by Levin and Hui-Chan [41]. This study examined measures to assess spasticity using spastic hemiparetic and normal adults as controls. Stretch reflexes (SR), H reflexes (i.e. tool to assess the modulation of spinal cord monosynaptic reflex activity [42]), and M responses (i.e. short latency muscle response to electrical stimulation [42]) of the soleus muscle with and without a high-frequency Achilles tendon vibration were assessed using EMG recordings from the soleus and tibialis anterior muscles. There was a significantly higher H/M ratio, shorter SR latencies, longer SR duration, and larger SR/M values in the hemiparetic group. Furthermore, the vibratory inhibition of the H reflex, which was previously found in studies with healthy adults [43], was not different between groups ($p > 0.05$). This study [41] attempted to explain the neural mechanism underlying how spasticity alters the muscle response to stretch reflexes and vibration. The authors hypothesized that the lower H reflex and SR latencies are caused by lower reflex thresholds in the patients with spasticity, leading to an earlier firing of motoneurons. If a lower reflex threshold were present, H reflexes would have shorter latencies because the stimulus would cause an earlier recruitment of motoneurons. Furthermore, more motoneurons would be recruited which would cause a larger response. Interestingly, the vibratory
inhibition of the H reflex was not different between groups. The finding suggests that there was not a deficit in presynaptic inhibition in the spastic hemiparetic patients. Alternatively, the authors suspected that the lack of a group difference was a result of higher inter-subject variability in the patients. Therefore, they suggested that a reduced reflex threshold might be present in those with spasticity.

Nardone and Schieppati [44] broke down latency responses in an attempt to explain reflex responses in patients with spasticity. This study examined short-latency and medium-latency responses in the soleus during evoked dorsiflexion with and without Achilles tendon vibration in spastic hemiparetic patients and normal adults as controls. The goal of this study was to determine how spindle group Ia and group II fibers contributed to abnormal reflex responses seen in spastic patients. One finding was that compared to a non-vibration condition, vibration decreased short-latency response by around 30% in normal adults ($p < 0.01$) but increased by 10% in the patients with spasticity in both legs. Nardone and Schieppati suggest that this is due to altered control over presynaptic inhibition on Ia terminals, which would be supported by previous findings that showed that patients with spasticity do not have a smaller H or tendon tap reflex when vibration is given. Another finding from this study was that vibration did not affect medium-latency response in normal adults but increased by 65% on the affected ($p < 0.001$) and 20% on the unaffected side of patients with spasticity when compared to a non-vibration condition. They suggest that this finding may have been a result of lower group II inhibition that is seen in normal adults. Lastly, during vibration without the evoked dorsiflexion, patients with spasticity had significantly higher muscle activity than they did without the vibration for both legs, which was not present in the normal adults. Nardone and Schieppati proposed that this
could be due to a combination of greater Ia input to soleus motoneurons that is then not counteracted by inhibition from group II input.

The theory behind the muscle tuning response being the mechanism behind an increase in muscle activity due to vibration comes from running studies [10, 27, 45, 46]. Evidence from these studies indicates that the body is able to tune its muscle activity as a way to reduce potentially harmful vibrations that could affect soft tissue in the body. This would happen through mechanical energy storage, where energy from the vibration is stored and returned from the muscle-tendon complex [10, 45]. Nigg [45] suggests that the muscle tuning response occurs right before heel strike during running, as a way to dampen impact forces, which acts as a vibration, from running. Wakeling et al. [46] tested this theory by mimicking the forces and vibration of forces from running on the foot using an actuator with frequencies between 10-65 Hz. Results from this study found that damping occurred in the soft tissues of the lower limb and resonance reduction occurred at heel strike but the frequency of the soft tissues still matched the input frequency of the vibrations, suggesting a muscle tuning response to the vibration by way of altering muscle activity. Both Wakeling et al. [46] and Rittweger [27] make note that vibration is transmitted from segment to segment and musculoskeletal stiffness could affect the amount of tuning and damping that occurs in response to vibration.

Where it is clear that certain types of vibration increases muscle activity in humans [28, 29, 31, 39, 40, 44], the mechanism in which this happens is not completely clear and may be from a combination of factors [10, 27, 41, 44, 45]. However, evidence suggests that a stretch reflex is involved to activate muscles [39, 40, 41, 44, 45], and spasticity and musculoskeletal stiffness in general may play a role
in the magnitude of response to vibration [27, 29, 41, 42, 46]. Furthermore, this response may act as a muscle tuning response to dampen forces acting on soft tissues in the body that are instead stored and released in the muscle-tendon complex [10, 27, 33, 45].

**Electromyography Methodology with Vibration**

When examining muscle activity in response to vibration at any frequency, many authors take note that the frequency of the vibration needs to be taken into account when analyzing electromyography (EMG) data [27, 39, 40, 46, 47]. Rittweger [27] notes that the vibration artifact needs to be accounted for in the EMG signal. Pollock et al. [39] also expresses the importance of accounting for vibration artifact in the EMG signal by filtering the signal to remove the artifact at the vibration frequency applied. Zaidell et al. [40] took this one step further and examined the frequency content of the signal in the EMG data in this study. Vibration artifact in the EMG signal was found at the vibration frequency as well as the harmonics of that frequency, giving further awareness of the need to remove these artifacts in the signal. A method of removing the frequencies and harmonics in the frequency content of the signal and replacing the removed content with the signal power of two neighboring frequencies was used to resolve the frequency artifact issue in this study.

Fratini et al. [47] specifically looked at the relevance of motion artifact in EMG signals during vibration. This study notes that during EMG recording, motion between electrodes and the skin and between skin layers causes motion artifact in raw EMG signals. Frequencies tested in this study ranged from 10-80 Hz, and male subjects were used to get muscle activity data from quadriceps muscles. Large amounts of artifact at the vibration frequency used as well as artifact at the harmonics
were found in the frequency content of the signal. Where Fratini et al. considered the amount of artifact that was found not to be negligible, this study recommended appropriate filtering techniques to remove vibration artifact from the frequency content of the raw EMG signal.

One study by Ritzmann et al. [48] suggests that some of what looks like artifact in the EMG signal may be actual muscle activity acting at the frequency of the vibration given. Methods in this study compared ankle ergometer evoked stretch reflexes and vibration evoked stretch reflexes by trying to separate stretch reflexes during vibration by putting a dummy electrode on the patellar tendon. Findings from this study suggest that vibration elicits stretch reflexes in the lower leg and that motion artifact is insignificant. Therefore, it is possible to use EMG data from vibration without needing additional filters. However, this study only used a vibration at a frequency of 5-30 Hz, and Fratini et al. [47] found that vibration artifact was less of a problem at lower frequencies by also putting an electrode on the patella. Therefore with slightly conflicting results from other studies, it is important to examine the frequency content of the vibration signal for each individual study to determine if significant motion artifact is present and should be removed [27, 39, 40, 47, 48].
Chapter 3
MANUSCRIPT

Introduction

Cerebral palsy (CP) is one of the most common causes of disability in children and is a neuromuscular disorder that limits mobility and motor control [1]. Poor muscle strength, balance and coordination associated with CP results in limited mobility and subsequent deficits in the musculoskeletal system [2, 4]. Therefore, it is critical to identify treatments that counteract the limited muscle and bone development in children with CP.

One treatment that has shown promise in offsetting the limited musculoskeletal development in children with CP is high-frequency, low-magnitude vibration (HLV). Studies in animals have reported a strong osteogenic effect of HLV, as demonstrated by improvements in bone formation rate, bone mineral density (BMD), trabecular structure, and cortical thickness [5]. Studies in humans are also promising [6, 7, 8] with observed improvements in the BMD of the tibia and spine [3] as well as greater increases in cortical bone area and general improvements in bone mass and muscle strength in children with CP [2, 22]. Similar findings have been reported in postmenopausal women [6, 7] as well as young women and adolescents with low bone mass [12].

Even though the effect of HLV on bone has been studied under a number of conditions with a variety of populations, the mechanism by which HLV acts on the musculoskeletal system is still relatively unknown [9]. One theory is that HLV acts
directly on bone cells through reactionary forces produced by the skeleton. Another theory is that the effect of HLV on bone results from an increase in muscle activity. It is likely that a spinal reflex is responsible for vibration-induced muscle activity where HLV elicits an excitatory response to muscle afferents, which then activate motoneurons [10, 27, 39, 40, 44]. This is supported by findings from Zaidell et al. [40], in which muscle activity increased during vibration in a loaded and unloaded leg, suggesting a spinal reflex mechanism. It is also possible that muscle activity occurs due to a muscle tuning response, where muscle activity is modulated in order to reduce the vibrations that pass through soft tissues in the body [10, 27, 45, 46].

If HLV does elicit a muscle response, the response may be exaggerated in children with spastic CP. Findings in a hemi-spastic stroke population supports this idea, where the affected side in adults who were hemi-spastic had a quicker rise to and larger peak force than the unaffected side [29]. Furthermore, studies examining altered reflex responses in spasticity suggest greater motoneuron excitation and lower inhibition to reflex responses elicited during vibration in hemiparetic stroke patients with spasticity [41, 44]. If the muscle response to HLV is greater in children with CP than in typically developing children, it may help explain the adaption of bone to HLV in disabled populations [2, 22] but not in healthy populations [30].

The purpose of this study was to determine if an HLV stimulus emitted by a floor-based platform during standing increases muscle activity in the lower extremities in children with spastic CP and if the increase is greater than in typically developing (TD) children. We hypothesized that muscle activity would increase in both children with spastic CP and TD children, but that the increase would be greater in the children with spastic CP.
Methods

Participants

Ambulatory children with CP and typically developing children, ages 4-11, were recruited for this study. Inclusion criteria for children with CP were having spastic diplegic or hemiplegic CP and a gross motor function classification of I-III. Inclusion criteria for typically developing children were being between the 5th and 95th age-based percentiles for height and body mass, matched to children with CP for age, sex and race, and taking no medications known to affect musculoskeletal development. Participants were excluded from this study if they had Botox treatment or surgery within the 12 months prior to the study. Children with CP were recruited from the AI duPont Hospital for Children and other children’s hospitals. TD children were recruited from the University of Delaware and the Newark, DE communities. The institutional review boards at AI duPont Hospital for Children and at University of Delaware approved this protocol (Appendix B). Legal guardians provided written informed consent and participants provided written informed assent if they were able.

Anthropometrics

Standing height was assessed using a stadiometer (Seca 217; Seca GmbH & Co. KG., Hamburg, GER), and body mass was assessed using a scale (Detecto D1130; Detecto, Webb City, MO) while children wore minimal clothing and were not wearing shoes or braces.
Tanner Staging

A physician assistant assessed the Tanner stage of each participant to determine sexual maturity. Tanner staging is assessed on a rating scale from 1 to 5, with 1 indicating no sexual maturity and 5 indicating sexual maturity [49, 50].

Gross Motor Function Classification System (GMFCS)

A physician assistant assessed gross motor function using the GMFCS. The GMFCS ranges from I to V, with I indicating impairments in some gross motor skills and V indicating no independent mobility [15]. Details on this scale can be seen in Appendix A.

Modified Ashworth Scale (MAS)

The MAS was used to assess plantar flexor tightness in participants with CP while the participant was lying supine on a table with the grade for each limb based on an average of three trials. The MAS grade is assessed on a scale from 0 to 4, with 0 indicating presence of normal muscle tone and 4 indicating muscle rigidity during plantar and dorsi flexion [51].

Instrumentation

Surface electromyography (EMG) signals were collected using Motion Lab Systems, Inc. (Baton Rouge, LA) hardware and bipolar surface electrodes that were a fixed 2 cm apart. The sampling frequency used was 2000 Hz and DASYLab Software (Measurement Computing Corp., Norton, MA) was used for data acquisition. A specifically designed MATLAB (Mathworks, Natick, MA) program was used to analyze the EMG signals. The HLV platforms used in this study were modified medical devices from Juvent Medical (Riveria Beach, FL). The devices emitted an
HLV of 30 Hz at 0.3 g, which is within the range of frequency and magnitude of vibration to be considered an HLV stimulus. A Biodex dynamometer was used to assess maximum voluntary isometric contractions (Biodex Medical Systems, Shirley, NY).

Data Collection

Data collection took place at University of Delaware. EMG was collected from six muscles of the lower extremity in the most affected limb of children with CP and of the nondominant limb of TD children. The skin was cleaned and slightly abraded with alcohol wipes prior to electrode placement. The tibialis anterior (TA), lateral gastrocnemius (LG), medial gastrocnemius (MG), soleus (SOL), biceps femoris (BF) and vastus lateralis (VL) muscles were examined in this study. Electrodes were placed based the Surface Electromyography for the Non-Invasive Assessment of Muscles project (SENIAM) recommendations for sensor locations on individual muscles [52] with the ground electrode placed on the patella.

Muscles were tested for validity in placement. To test for TA electrode placement, participants were asked to dorsi flex against the force of the tester’s hand on the top of the foot. To test for LG, MG, and SOL placement, participants were asked to plantar flex against the tester’s hand on the underside of the foot. Participants were asked to kick forward at the ankle against the force of the tester’s hand on the anterior side of the ankle to check for VL electrode placement. Similarly, participants were asked to pull their leg back, bending at the knee, against the force of the tester’s hand on the posterior side of the ankle to test for BF electrode placement. During these tests, the EMG system was also adjusted for gains to get optimal signal acquisition.
The individual gain for each muscle was increased until the signal was noticeable but not outside the range of measurement in the data acquisition program.

Participants stood on the HLV platform independently and without support during the HLV treatment. The set-up of a child standing on the HLV platform can be seen in Figure 1. The HLV treatment consisted of three different conditions that were 30 seconds each. In the first condition the platform was off (i.e., pre-HLV). In the second condition the platform was on (i.e., HLV). In the third condition the platform was off again (i.e., post-HLV). The post-HLV condition was used to determine that any changes seen between the pre-HLV and HLV conditions were due to vibration and not to prolonged standing. Participants were asked to stand with their knees as straight as possible, their feet as flat as possible and facing forward while standing as still as possible for all conditions. A research assistant stood on either side of the participant to prevent falls. A wooden chair was placed in front of the HLV platform for support, if needed. However, participants were instructed to stand without support. It was documented when a participant leaned on the chair, swayed or moved noticeably while standing on the platform. Only data collected during independent standing was used for the analysis.
Electrodes remained in the same place, and participants were asked to do a maximal voluntary isometric contraction (MVIC) on a Biodex dynamometer. MVIC data was collected from the most affected limb in children with CP and the nondominant limb of TD children. All children were strapped across the chest, waist, and thigh, and knee flexion was kept as close to zero as possible but within 20 degrees of flexion. The protocol for MVIC collection was 5 seconds of maximal plantar flexion followed by 30 seconds of rest and then 5 seconds of maximal dorsiflexion followed by 30 seconds of rest. This MVIC protocol was repeated three times.
Data Analysis

The EMG data was processed through a specialized MATLAB program coded to analyze the data from this study. A correction for gain adjustments was made to account for the potentially different gain adjustments during data collection. A high pass, $20 \text{ Hz}$, 4th order Butterworth filter and low pass, $500 \text{ Hz}$, 4th order Butterworth filter were used to filter the raw signal. A frequency filtering technique was used to filter out artifacts at the frequency of the vibration and the corresponding harmonics of that frequency, as recommended by Fratini et al. [47]. This technique transformed the EMG data from the time domain to the frequency domain, and a 5 Hz-sized window passed through the data. A threshold value was set at three times the median frequency of the segment of data in each window. If any data point was above the threshold value, it was replaced with a local median. The data was then transformed back into the time domain for further processing. Details of this filtering technique are presented in Appendix C.

The root mean square (RMS) amplitude values with a one second window were calculated from the 30 seconds of data for pre-HLV, HLV, and post-HLV conditions for each of the six muscles. For the analysis of MVIC data, the plantar flexion MVIC that created the greatest plantar flexion torque was used for analysis of EMG data for the LG, MG, and SOL only. The dorsiflexion MVIC that created the greatest dorsiflexion torque was used for analysis of the EMG data for the TA. The RMS amplitude values were calculated for each of the four muscles assessed for MVIC with a 0.5 second window around the maximum torque produced. The RMS values within that window were calculated for both plantar flexion and dorsiflexion actions to determine if any co-contraction was present. Only plantar flexion and
dorsiflexion MVIC’s with minimal activity seen in the other muscles not being targeted for MVIC were used for analysis.

Change in muscle activity between conditions was assessed for each of the six muscles. Muscle activity for each condition for the TA, LG, MG, and SOL muscles were normalized to muscle activity obtained from MVIC data to further assess the change in muscle activity between conditions in this study.

Statistical Analysis

Within-subject and between-group comparisons were made in this study. Using SPSS (IBM Corp., Armonk, NY) software and repeated measures ANOVA, differences between pre-HLV, HLV, and post-HLV conditions and differences between groups were determined. Significance level was set at $p < 0.05$. The size of the effects was determined using partial eta$^2$.

Results

Eleven children with CP (7 boys and 4 girls) and 10 TD children (7 boys and 3 girls) participated in this study. Participant descriptives are presented in Table 1. There were no significant differences between groups in any of the measured anthropometrics or sexual maturity. However, the TD children could produce a significantly greater MVIC than children with CP ($p = 0.001$). There were no significant differences between pre-HLV and post-HLV conditions in children with CP or TD children as seen in Figure 2. Therefore, to limit the number of comparisons, only the pre-HLV condition was compared to the HLV condition. Results with differences between when vibration is applied (HLVon) and standing with no vibration (HLVoff) are presented in Figure 3. There was a significant condition effect
with an increase in muscle activity due to vibration in the MG \((p = 0.029, \text{ partial } \eta^2 = 0.228)\), SOL \((p = 0.029, \text{ partial } \eta^2 = 0.226)\), and BF \((p = 0.011, \text{ partial } \eta^2 = 0.296)\) muscles in the combined TD and CP groups. There was a significant group by condition effect in TA \((p = 0.033, \text{ partial } \eta^2 = 0.219)\), LG \((p = 0.002, \text{ partial } \eta^2 = 0.391)\), and VL \((p = 0.015, \text{ partial } \eta^2 = 0.274)\) with an increase in muscle activity in the HLVon versus the HLVoff condition in children with CP but not in TD children.
<table>
<thead>
<tr>
<th></th>
<th>CP (n = 11)</th>
<th>TD (n = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y)</td>
<td>8.9 ± 2.1</td>
<td>8.7 ± 1.7</td>
</tr>
<tr>
<td>Height (m)</td>
<td>1.24 ± 0.10</td>
<td>1.21 ± 0.42</td>
</tr>
<tr>
<td>Height (percentile)</td>
<td>19.7 ± 24.5</td>
<td>56.3 ± 31.7</td>
</tr>
<tr>
<td>Body mass (kg)</td>
<td>26.2 ± 6.8</td>
<td>29.8 ± 6.8</td>
</tr>
<tr>
<td>Body mass (percentile)</td>
<td>29.6 ± 29.7</td>
<td>57.8 ± 26.7</td>
</tr>
<tr>
<td>BMI (kg/m$^2$)</td>
<td>16.7 ± 2.7</td>
<td>16.9 ± 2.9</td>
</tr>
<tr>
<td>BMI (percentile)</td>
<td>48.4 ± 32.1</td>
<td>48.4 ± 33.5</td>
</tr>
<tr>
<td>Tanner Stage (1 / 2 / 3)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pubic hair</td>
<td>6 / 4 / 1</td>
<td>9 / 1 / 0</td>
</tr>
<tr>
<td>Breast/testicular</td>
<td>8 / 2 / 1</td>
<td>7 / 2 / 1</td>
</tr>
<tr>
<td>GMFCS (I / II / III)</td>
<td>5 / 4 / 2</td>
<td>N/A</td>
</tr>
<tr>
<td>MAS</td>
<td>1.82 ± 0.81</td>
<td>N/A</td>
</tr>
<tr>
<td>MVIC (N•m)</td>
<td>8.17 ± 7.24 *</td>
<td>36.50 ± 16.72 *</td>
</tr>
</tbody>
</table>

Table 1 Subject descriptors for both CP and TD groups are presented, including anthropometrics, Tanner stage, Gross Motor Function Classification System (GMFCS), Modified Ashworth Scale grade (MAS), and maximal voluntary isometric contraction torque (MVIC). MVIC was the only variable significantly different between groups. * $p < 0.05$. 
Figure 2  Differences between the HLV off condition prior to HLV (PreHLVoff) and the HLV off condition after HLV (PostHLVoff) are presented for each muscle for A: children with CP and B: TD children. There were no significant differences between the two conditions for any muscle in either group.
Figure 3  Mean differences between HLVon and HLVoff are presented for the tibialis anterior (TA), lateral gastrocnemius (LG), medial gastrocnemius (MG), soleus (SOL), biceps femoris (BF), and vastus lateralis (VL).

*Condition effect with HLVon > HLVoff for the cerebral palsy (CP) and typically developing (TD) groups combined, $p < 0.05$. **Group by condition effect with HLVon > HLVoff in the CP group but not in the TD group, $p < 0.05$.

There was a significant condition effect with an increase in muscle activity due to vibration when normalized to MVIC in the LG ($p = 0.044$, partial eta$^2 = 0.196$) muscle in the combined TD and CP groups. There was a significant group by condition effect in TA ($p = 0.035$, partial eta$^2 = 0.209$) muscle with an increase in muscle activity in the HLVon versus the HLVoff condition in children with CP but not in TD children.
Discussion

Results from this study support the hypothesis that a floor-based HLV stimulus increases muscle activity in the lower extremities of children with spastic CP. There were no differences in muscle activity before and after the HLV stimulus was applied in either group, which confirms that the increase in muscle activity during the HLV condition was due to the HLV treatment and not to prolonged standing. Additionally, when normalizing EMG data from the vibration conditions to an MVIC for the plantar and dorsi flexors used in this study, results were consistent with results from the non-normalized data. This further supports the hypothesis that HLV increases muscle activity increases in children with spastic CP. Findings from this study also help to describe a potential mechanism for musculoskeletal response to HLV, where improvements in bone in disabled populations [2, 3, 22] but not in healthy populations [30] could partly be described by greater vibration-induced muscle activity in those with spastic CP.

These results indicate that the muscle response to HLV is exaggerated in children with spastic CP as compared to TD children. This supports the conclusion by Suresh et al. [29] that spastic muscle is more sensitive to vibration than healthy muscle. Muscles in children with spastic CP, such as those included in this study, are more sensitive to stretch [24]. A previous study by Levin and Hui-Chan [41] examined the H reflex and stretch reflex response with and without Achilles tendon vibration in adults with and without hemiparesis and spasticity. The authors found reduced H and stretch reflex latencies and longer stretch reflex durations in the patients with spasticity and suggested that this may be due to reduced reflex thresholds in those with spasticity. If this were the case, motoneurons would be recruited earlier and in greater
numbers from an electrical stimulus causing a larger response and short H reflex latencies as they saw in this study. Nardone and Schieppati [44] further investigated the potential reflex pathways associated with spasticity in hemiparetic patients with spasticity and healthy adults. Foot dorsiflexion perturbations were given with and without Achilles tendon vibration in an attempt to identify the contribution of spindle group Ia and II fibers to reflex responses in patients with spasticity. This study found that vibration decreased the short-latency response in normal subjects but increased the short-latency response in patients with spasticity. They attributed this phenomenon to altered control of presynaptic inhibition on Ia terminals, which was supported by previous studies that found that patients with spasticity don’t have reduced H reflexes with vibration, which is a phenomenon that occurs in healthy adults [43]. Nardone and Schieppati [44] also found no effect on the medium-latency response in healthy adults but an increased medium-latency response in healthy patients with perturbations given during vibration. They suggest that this could be due to reduced group II inhibition. Furthermore, during vibration without perturbation, patients with spasticity had significantly greater muscle activity than during non-vibration. They theorize that these results may be due to a combination of increased Ia input to the soleus muscle and decreased group II inhibition that would normally counteract the input. Though previous findings [41, 44] were based on Achilles tendon vibration in an attempt to understand reflex response in hemiparesis patients with spasticity, it is possible that children with spastic CP also have this highly sensitive spinal reflex response that induces greater muscle activity during vibration than that of TD children.

The comparison of the HL Von and HL Voff conditions when normalized to MVIC gives insight into the degree to which HLV increases muscle activity in
children with CP. The increase in muscle activity during HLV was approximately 8% in the TA and approximately 15% in the LG, with the MG and SOL also greater than the TD children, although the differences were not significant. The 8-15% increase in muscle activity associated with HLV in the children with CP may seem modest; however, the increase is beyond the muscle activity associated with standing and would occur throughout a 10-20 minute session, as this was the time previously used to assess the effect of HLV on muscle and bone in previous studies [2, 3, 22]. Furthermore, a previous study [54] that examined individual muscle contributions to contact force during walking in healthy adults found peak contributions throughout the gait cycle were around 30% for the TA, 60% for the MG, 40% for the SOL, 15% for the BF, and 25% for the vastus medialis. Since these increases in muscle activity during walking in healthy individuals are not near maximal contribution during walking even at their own individual peaks during the gait cycle and would only occur for a fraction of a second over each gait cycle, it helps put into perspective the consistent increase in muscle activity with HLV seen in children with CP in this study.

Children with CP and TD children were matched for age, and there was a similar composition of sex and race, which was a major strength of this study as differences in musculoskeletal properties that may exist due to these variables were minimized. Furthermore, there were no significant differences between groups in any measured anthropometrics, including height, body mass, and BMI, and there were no differences in age or sexual maturity level. The similarity of the groups reduces the chance that any differences in muscle response seen between groups could have been due to other extraneous variables. An additional strength of this study was that all children were able to stand independently. Therefore, any potential influences of
compensatory mechanisms needed to remain upright were minimized. Another notable strength of this study was the unique filtering technique used during data processing. As noted by Fratini et al. [47], many previous studies have not taken into account motion artifact at the frequency of the vibration and corresponding harmonics. However, findings from the study by Fratini et al. and a spectral analysis of raw data from this current study clearly indicated a need to eliminate vibration-induced motion artifact while minimally reducing content of the EMG signal representing muscle activity. Data processing using the filtering technique in this study provides confidence that changes in muscle activity due to vibration were not misrepresented.

One limitation in this study was that only children with mild to moderate CP were used, meaning that this population of children with CP represents some of the more independently mobile cases of those with CP. Most children had a GMFC of I or II, and those with a GMFC of III could stand independently for the duration of the vibration protocol. Future studies could benefit a larger population of those with CP by examining the effects of vibration under different loading conditions to determine if a similar muscle response occurs when independent standing on a platform is not an option due to physical disabilities. Another limitation was that posture was not controlled for in this study. Children with spastic CP often stand differently from each other as well as from typically developing children due to muscle tightness associated with spasticity. Therefore, there was the limitation of the inability to fully control for posture. However, since children with mild to moderate CP participated in this study, differences in posture were minimized. This study also did not look at MVIC normalized values for all muscles tested. This was a limitation of the protocol used in this study and muscle tightness associated with a population with spastic CP.
Furthermore, it has been reported that children with CP have greater variability in voluntary contraction tasks compared to typically developing children [53]. However, knee angle was kept between zero and 20 degrees of flexion when obtaining MVIC, which is very close to the position of the leg during standing. Therefore, electrode placement over the muscle between conditions and leg position when standing and the MVIC condition seated on the Biodex dynamometer are very similar. Additionally, three MVIC attempts were performed for both plantar flexion and dorsiflexion, and only the best attempt was used for normalization. Therefore, it was assumed that an accurate representation of a MVIC was used for normalization. Future studies could benefit from examining differences between a standing and vibration condition by normalizing to MVIC for even more muscles or normalizing to a submaximal contraction. Lastly, a specific floor-based HLV of 30 Hz at 0.3 g was used in this study, so findings cannot be generalized to other types of vibration. However, this specific level of vibration was used because it was determined to be safe as well as effective in showing improvements to the musculoskeletal system in previous studies [2, 6, 31]. Where surgery and invasive methods are often used in the treatment of children with CP [25, 26], HLV has the benefit of being a noninvasive and non-pharmacological treatment to improve musculoskeletal health in children with CP.

**Conclusions**

In conclusion, HLV during standing introduces a mechanical load to the body, activating muscles. However, children with spastic CP have a greater muscle response to vibration than their typically developing peers. The exaggerated muscle response in children with CP may be due to a highly sensitive reflex response in children with spastic CP [29, 41, 44]. However, additional studies are needed to confirm this notion.
Results from this study suggest that the greater muscle response to HLV may play a role in the adaptation of the musculoskeletal system to HLV in disabled populations [2, 22] but not in healthy populations [30]. Findings from this study are important, as they support the idea that HLV could serve as a noninvasive, non-pharmacological approach to improving musculoskeletal health in children with CP.
REFERENCES


55. Burden A. How should we normalize electromyograms obtained from healthy participants? What we have learned from over 25 years of research. *J Electromyogr Kines* 2010;20:1023-1035.


Appendix A

GROSS MOTOR FUNCTION CLASSIFICATION SCALE

Table I: Description of gross motor function for children aged 6 to 12 years by GMFCS level (Palisano et al. 1997)

<table>
<thead>
<tr>
<th>Level</th>
<th>Expected gross motor function between age 6 and 12 y</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>Children walk indoors and outdoors, and climb stairs without limitations. Children perform gross motor skills including running and jumping, but speed, balance, and coordination are reduced.</td>
</tr>
<tr>
<td>II</td>
<td>Children walk indoors and outdoors, and climb stairs holding onto a rail, but experience limitations walking on uneven surfaces and inclines, and walking in crowds or confined spaces. Children have at best only minimal ability to perform gross motor skills such as running and jumping.</td>
</tr>
<tr>
<td>III</td>
<td>Children walk indoors or outdoors on a level surface with an assistive mobility device. Children may climb stairs holding onto a rail. Depending on upper-limb function, children propel a wheelchair manually or are transported (pushed by another person) when travelling for long distances or outdoors on uneven terrain.</td>
</tr>
<tr>
<td>IV</td>
<td>Children may maintain levels of function achieved before age 6 years or rely more on wheeled mobility at home, school, and in the community. Children may achieve self-mobility using a powered wheelchair.</td>
</tr>
<tr>
<td>V</td>
<td>Physical impairments restrict voluntary control of movement and the ability to maintain antigravity head and trunk postures. All areas of motor function are limited. Functional limitations in sitting and standing are not fully compensated for through the use of adaptive equipment and assistive technology. Children have no means of independent mobility and are transported (pushed by another person). Some children achieve self-mobility using a powered wheelchair with extensive adaptations.</td>
</tr>
</tbody>
</table>
Appendix B

INSTITUTIONAL REVIEW BOARD APPROVAL LETTERS

DATE: December 19, 2014
TO: Christopher Moddox, PhD
FROM: University of Delaware IRB
STUDY TITLE: [359757-4] Effect of botox and vibration on bone in children with cerebral palsy UD
SUBMISSION TYPE: Continuing Review/Progress Report
ACTION: APPROVED
APPROVAL DATE: December 19, 2014
EXPIRATION DATE: November 20, 2015
REVIEW TYPE: Administrative Review

Thank you for your submission of Continuing Review/Progress Report materials for this research study. The University of Delaware IRB has APPROVED your submission. All research must be conducted in accordance with this approved submission.

This submission has received Administrative Review based on the applicable federal regulation.

All serious and unexpected adverse events must be reported to the reviewing IRB.

Please report all non-compliance issues or complaints regarding this study to the reviewing IRB and this office.

Please note that all research records must be retained for a minimum of three years.

Based on the risks, this project requires Continuing Review is required on an annual basis.

If you have any questions, please contact Maria Pasazanos at (302) 831-6015 or mmarapzg@udel.edu. Please include your study title and reference number in all correspondence with this office.
DATE: February 27, 2012
TO: Freeman Miller, MD
FROM: Nemours Delaware IRB
STUDY TITLE: [115648-2] Effect of Botox and vibration on bone in children with cerebral palsy
RB #: 115648
SUBMISSION TYPE: Response/Follow-Up
ACTION: APPROVED
APPROVAL DATE: January 26, 2012
EXPIRATION DATE: January 26, 2013

Thank you for your submission of Response/Follow-Up materials for this research study. Your initial submission received full review at the January 26, 2012 meeting and met all DHHS [and FDA] criteria for approval. The approval was contingent on the response to minor stipulations. Your response has received Expedited Review and is accepted. The above-referenced research study is approved.

The IRB has determined that:

- This is "Research involving greater than minimal risk but presenting the prospect of direct benefit to the individual subjects per 45CFR46.405 and 21CFR50.52".
- Parental permission is required prior to initiation of any research procedures using only the most current IRB approved form(s) posted as a Board Document in IRBNet. All protocol documents, including board approved documents are found in the "Study Designer" for each study in IRBNet.
- The IRB approved telephone script is required prior to initiation of any research procedure.
- The permission of one parent is sufficient. A person who is not a parent may not give permission without prior IRB review and approval.
- Assent of minors is required prior to initiation of any research procedures, using only the most current assent form(s) posted as a Board Document in IRBNet. If the investigator chooses to obtain assent, the form(s) listed below must be used and the minor's dissent must be honored.
- A signed copy of the Parental Permission/Informed Consent form must be included in the Nemours' medical record. Research data may also be included into the Nemours medical record.
- To continue, the research requires IRB review and approval on an annual basis. Otherwise, January 25, 2013 is the last day that research may be conducted. The Principal Investigator is responsible for the timely submission of the continuing review application. Please post this date on your research calendar.

Reviewed/approved documents in this submission:

- Application Form - Revised Application Form (UPDATED: 02/14/2012)
- Child Assent - Revised child assent (UPDATED: 02/14/2012)
The IRB requires that a copy of the participant brochure: "Becoming A Research Volunteer" will be given to every individual enrolled in a research study. The PDF file for this document has been attached to this study as a Board Document.

**Investigator Agreement:** As the PI, you have agreed to assure that this research is conducted in compliance with Nemours policy and all applicable federal regulations and ICH standards, including, but not inclusive of:

- All research must be conducted in accordance with its approved submission. Any revision to approved materials must be approved by the IRB prior to initiation.
- Remember that informed consent/parental permission is a process beginning with a description of the study and insurance of participant understanding followed by a signed consent form. Informed consent must continue throughout the study via a dialogue between the researcher and research participant. Federal regulations require each participant receive a copy of the signed consent document.
- All serious and unexpected adverse events and unanticipated problems affecting participants must be reported promptly to the IRB according to NOHSP policy.
- All non-compliance issues or complaints regarding this study must be reported to the Director, NOHSP.
- All research records must be retained for a minimum of three years.
- A Closure Report must be submitted to the IRB when this protocol is completed.

If you have any questions, please contact Camille Varacchi at Nemours Al duPont Hospital for Children 1600 Rockland Road, ARB-Room 201, Wilmington, Delaware 19803, 302-661-6807 or varacchi@nemours.org.

Please include your study title and reference number in all correspondence with this office.
Appendix C

FREQUENCY FILTERING TECHNIQUE

Based on a study and findings by Fratini et al. [47], a frequency analysis was performed on vibration data in this study. Non-physiological spikes of data around the vibration frequency and its harmonics were seen in the frequency analysis of all vibration trials in this study. An example of this frequency pattern from one muscle from one vibration condition can be seen in Figure 4.

![Frequency spectrum from the LG of a vibration trial where the unfiltered data is represented in black and the filtered data is overlaid represented in grey.](image)

Figure 4  Frequency spectrum from the LG of a vibration trial where the unfiltered data is represented in black and the filtered data is overlaid represented in grey.

Due to the spikes seen in the frequency, a filtering method was applied. For this analysis, a Fast Fourier Transform was performed and the data was transformed from the time domain to the frequency domain. Then a 5 Hz sized window passed
through the data. If any data points were greater than three times the standard deviation of the data within each window, then those data points were removed and replaced with a local median. All data was then transformed back into the time domain for further analysis. An example of how EMG data looked before and after filtering can be seen in Figure 5.

Figure 5  EMG data from the LG of a vibration trial where the unfiltered data is represented in black and the filtered data is overlaid represented in grey.