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Patterned Plantar Stimulation During Gait

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Patterned Plantar Stimulation During Gait

A Thesis

Presented to the Department of Biomechanics

and the

Faculty of the Graduate College

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In Partial Fulfillment

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by

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Spring 2023

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Patterned Plantar Stimulation During Gait

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It is well established that the soles of the feet are involved and aid in balance control. However, it is not well understood the exact role that the feet play in gait control. During walking, the center of pressure (CoP) takes a predictable and repeated path along the plantar surfaces, going from heel to toe. This CoP has been established to be vital for postural control during standing, the plantar surfaces may perform a similar role during walking by perceiving this CoP path. Most studies use vibro-tactile stimulation on the plantar surfaces during the entire gait cycle, including the swing phase. However, no studies have investigated the effects of different patterns of sequential stimulation on the plantar surfaces during the stance phase of gait. Therefore, the following chapters describe a method of testing this effect, and demonstrating how such patterned plantar stimulation alters gait in healthy young adults. This method of testing was developed such that plantar stimulation would activate specifically during the stance phase of the gait cycle, and activate in a gait-like or an abnormal sequence. We then hypothesized that stimulation in an abnormal sequence would result in gait and balance deficits when compared to stimulation that followed the natural sequence during walking. Additionally, that walking on an inclined surface would increase the effects of the tactile stimulation sequences on such measures when compared with no stimulation. We tested a total of

nine healthy adults and found very minimal effects from the stimulation in any pattern. This demonstrates that healthy adults have the ability to adjust and reweigh sensory information from the plantar surfaces such that gait and balance outcomes show minimal or no deficits when foot-sole tactile sensory sequences are manipulated during slow walking. Additionally, that the perception of the CoP movement may be predominately supplied by slow adapting fibers that are not typically sensitive to vibrations. This work gives indication to the flexibility and adaptability of a healthy motor control system and demonstrates a method of testing such a system with an online stimulation control software.

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Symbols and Abbreviations

CoP	Center of Pressure
CNS	Central Nervous System
CoM	Center of Mass
BOLD	Blood Oxygen Level Dependent
MRI	Magnetic Resonance Imaging
FA	Fast Adapting
SA	Slow Adapting
EMG	Electromyography
XCoM	Extrapolated Center of Mass
MoS	Margin of Stability
BoS	Base of Support
PWS	Preferred Walking Speed
SOT	Sensory Organization Test
SMA	Supplementary Motor Areas
M1	Motor Cortex
VLSM	Voxel-Based Lesion Symptom Mapping
MT	Metatarsal Joint
NS	No Stimulation
GS	Gait-Like Stimulation
RS	Random Stimulation
AP	Anteroposterior
ML	Mediolateral
GRF	Ground Reaction Forces

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Chapter 1: Introduction

It is well established that sensory information from the cutaneous mechanoreceptors within the plantar surfaces of the feet are important for the control of standing and walking balance (Inglis et al. 2002; Felicetti et al. 2021). Specifically, they aid in the perception of changes to the walking environment (Maurer et al. 2001) by responding to changes in pressure, stretch, and vibrations on the skin (Inglis et al. 2002; Kavounoudias et al. 1998). This information is important in perceiving the body's orientation in space (Kavounoudias et al. 2001) through analyzing the position of the center of pressure (CoP) on the sole of the foot with respect to the rest of the body (Roll et al. 2002). However, during walking, this CoP traverses along the foot and is adjusted during gait to remain in balance (Hoff et al. 2007). Thus, the movement of this CoP would be vital to the central nervous system (CNS) to maintain balance during walking.

Taking this line of thinking even further, it is a possibility that the CNS is specifically sensitive to this pattern of movement the CoP exhibits on the plantar surfaces during normal walking. Interestingly, the arrangements of specific fast adapting tactile receptors within the plantar surfaces show increased sensitivity to a moving stimulus as opposed to a static stimulus (Stzalkowski et al. 2018). It has been shown that when specific regions of the plantar surface were desensitized, the CoP during walking was shifted away from the regions of desensitization and towards regions that remained sensitive (Nurse & Nigg 2001). Thus, the arrangement of tactile receptors on the plantar surfaces could be important for perceiving the movement of the CoP. These receptors were also shown to have an instantaneous effect on gait, when stimulating regions of the

foot during different phases of the gait cycle, different muscle responses occurred (Zehr et al. 2014). Therefore, stimulating the plantar surfaces could augment the perception of the CoP movement pattern.

The CoP is a common dependent variable that is investigated for determining balance control during standing and walking (Hof et al. 2007, Kavounoudias et al. 1998, Meyer et al. 2004, Nurse & Nigg 2001). This is because it is the point at which all the pressure is directed towards the ground, and its location can have a strong effect on the movement of the center of mass (CoM) (Hof et al. 2007). This is especially true during inclined walking where the risk of a slip or fall is much greater than in level walking due to an increase in shear forces (Hanson et al. 1999, Sun et al. 1996). With the plantar surfaces of the feet being the only surface in contact with the support surface, it could play an even more important role in balance control during inclined walking by sensing the changing position of the CoP (Hanson et al. 1999, Viseux 2020). Based on the importance of the CoP, it would make sense, from an evolutionary perspective, to have a system that is sensitive to receiving and interpreting such information for proper balance control during a dynamic task such as walking. Therefore, having the ability to stimulate the plantar surfaces in a manner that can positively or negatively affect the perception of this CoP movement could increase or decrease balance control, respectively, during more complex walking tasks especially in those who have perceptual deficits.

Brain imaging studies have found a strong link between plantar surface stimulation and motor control centers when measuring Blood-oxygen-level dependent (BOLD) responses with Magnetic Resonance Imaging (MRI) (Zhang et al. 2019, Labriffe et al. 2017). Imagining walking tasks have been shown to activate similar brain centers

that also activate from simply stimulating the plantar surfaces in a gait-like manner (Labriffe et al. 2017). Interestingly, stimulating the surface of the hand in a sequence at various speeds led to different levels of brain responses (Oh et al. 2017). Specifically, there was an increase in brain activity when the hand was stimulated at a speed of 25cm/s and decreased when stimulated at 60cm/s (Oh et al. 2017). If this phenomenon is present within the hand, it would make sense for the foot to have a particular sensitivity to speed as well. Especially when there is a stimulus that moves along the foot every step and gives vital information about controlling balance during gait: the CoP. It could be that the CNS pays attention to this CoP movement pattern and speed to control balance during gait.

What would happen if the interpretation of this movement of the CoP was to be affected? One clinical population that could aid in answering this question is stroke survivors. Controlling balance during gait requires actively gaining and interpreting redundant sensory information coming from the visual, vestibular, and somatosensory systems (Mackinnon 2018). This redundancy allows for adaptability and compensations when one or multiple of the sensory modalities are impaired (Mackinnon 2018). However, if a lesion has occurred in any of the regions that are involved in the processes of sensing, interpreting, or outputting motor commands, major impairments to movement control can be the result (Mackinnon 2018). This can be seen as a decrease in walking speed, asymmetric walking patterns, and decrements in dynamic balance control in chronic stroke survivors (Forster & Young 1995; Mayo et al. 1999). Therefore, stroke survivors may be unable to interpret the movement of the CoP across the foot for proper balance control during gait.

There seems to be an important connection between sensation and motor recovery for stroke survivors. Previously, it was found that increased ipsilesional BOLD responses in the sensorimotor cortex from tactile stimulation was directly related to motor recovery of the paretic upper limb (Schaechter et al. 2012). This could be demonstrating a property of stroke recovery known as reorganization. This phenomenon is also known as relearning, where the individual must relearn how to perform tasks that the lesioned area was involved in before the stroke (Grefkes & Ward 2014). Thus, stimulating the plantar surfaces in a manner that follows the normal CoP pattern across the foot could bring more attention to the speed and pattern of movement of the CoP while walking, and allow for increased feedback about the environment. This increased feedback could help relearn the natural and sequential gait pattern.

Therefore, creating a live control system that can apply vibro-tactile stimulation either following the normal CoP path or going against that path would allow us to augment the perception of this CoP movement during each individual step. Then, investigating the effects of such augmentation during gait in healthy individuals would give indications of how altering the perception of the path the CoP travels during walking may affect balance control.

Purpose

The purpose of this study will be to determine how different patterns of sensory stimulation to the plantar surfaces of the feet alters the control of walking behavior in healthy individuals.

Aims & Hypotheses

Aim 1: Develop a live vibro-tactile control system that applies vibrations to different regions of the plantar surfaces that either follows the normal CoP path or goes against that path during the stance phases of gait.

Aim 2: Determine how different patterns of vibro-tactile stimulation to the plantar surfaces affect spatiotemporal and balance measures of gait in healthy adults.

Hypothesis 2a: Vibro-tactile stimulation in an abnormal pattern (random stimulation) during gait, will result in gait (stance times, stance lengths) and balance deficits (stride width, foot placement, and margins of stability) when compared to stimulation that follows a natural walking pattern.

Hypothesis 2b: Walking on an inclined surface will increase the effect of tactile stimulation on balance and gait measures than walking without tactile stimulation.

Chapter 2: Literature Review

Section 1: An Anatomical View

Cutaneous Receptors

To understand how stimulating or augmenting the cutaneous receptors in the foot might affect gait and balance control, it is first important to understand what these cutaneous receptors are and how they might interact with the rest of the body. Cutaneous mechanoreceptors are the “fundamental units for the transduction and transmission of tactile feedback to the CNS” (Stzalkowski et al. 2018). They respond to deformations of the skin caused by either vibrations, pressure, or stretching. Four different nerve fibers have been established that exhibit unique firing characteristics: Two types of Fast adapting fibers (FAI & FAII) and two types of Slow adapting fibers (SAI & SAI) (Stzalkowski et al. 2018). FA afferents are more sensitive to mechanical stimuli that demonstrate a rate of change and cease firing once the stimulus is static or sustained, with the removal of the stimulus causing the firing of these fibers once again. While the opposite occurs for SA afferents; they continue to fire throughout sustained stimulation and are not as sensitive to changing stimuli. The types of these different fibers are distinguished by their receptive fields (Stzalkowski et al. 2018). Type I fibers have small receptive fields, within the foot being about 78mm^2 , and type II fibers have larger receptive fields, about 560mm^2 (Stzalkowski et al. 2018). There is a specific distribution of these fibers as well, within the plantar surface the majority of the fibers, about 60%, are FA fibers (48% type I and 13% type II), with the remaining being SA fibers (18% type I and 21% type II) (Stzalkowski et al. 2018). This suggests that the foot is

anatomically wired to be sensitive to changes of stimuli (FA fibers) and specific locations (FAI) of stimuli, such as pressures, on the foot sole.

It is important to go into further detail about the actual mechanical units that these nerve fibers innervate to understand the type of stimuli they respond to best. These mechanoreceptors are the functional units altering physical deformations to action potentials (Zimmerman et al. 2014). There are four different types of mechanoreceptors these nerve fibers innervate: Merkel cells for SAI fibers, Ruffini corpuscles for SAII, Meissner's corpuscles for FAI, and Pacinian corpuscles are innervated by FAII fibers (Zimmerman et al. 2014). These mechanoreceptors are specialized to respond to varying types of stimuli applied to the skin. Merkel cells are sensitive to static indentation of the skin, while Ruffini corpuscles are sensitive to the stretch of the skin (Zimmerman et al. 2014). With these mechanoreceptors being innervated by slow adapting fibers, the signals from these fibers continue during the duration of stimulation. On the other hand, Meissner's corpuscles and Pacinian corpuscles respond predominantly to movement and vibrations applied to the skin (Zimmerman et al. 2014). Specifically, Meissner's being sensitive to vibrations below 40Hz, and Pacinian's being sensitive to higher frequencies of around 200Hz (Zimmerman et al. 2014). These later two mechanoreceptors would be the main sources of afferent information due to vibro-tactile stimulation on the plantar surfaces, however the SA innervated mechanoreceptors may also be affected by such stimulation (see *Stimulation During Standing*).

Due to these mechanoreceptors responding to varying types of stimuli, they respectively give different information about the environment they are placed in. Firstly, Merkel cells respond at different levels based on varying levels of pressure, with an

increase in pressure the Merkel cell cause more frequent action potentials (Zimmerman et al. 2014 & Gardner 2010). Therefore, these Merkel cells give indication to how much pressure is being applied to respective regions of the skin. This could be particularly useful for the perception of the CoP. Secondly, Meissner corpuscles respond to initial contact, to motion across the skin, and low frequency vibrations (Zimmerman et al. 2014; Gardner 2010). This would apply to a stimulus sliding or slipping across the surface of the skin. Therefore, Meissner corpuscles could be one of the major mechanoreceptors used to alert the body of a possible slip along the plantar surface during walking. There's a strong possibility that these receptors could still be used to detect a slip when wearing shoes, possibly by sensing the resonant vibrations that occur when sliding one surface (a shoe) across another surface (the ground). Finally, Pacinian corpuscles may be used to determine the rigidity of an object (Gardner 2010). Gardner uses the example of placing a hard block onto another hard object. This would cause high frequency resonant vibrations to travel across the block, which could be received by the Pacinian corpuscles and thus give indications of the rigidity of the object being held, as well as the other object that collided with the block (Gardner 2010). Therefore, these mechanoreceptors may be used to determine the rigidity of the environment that one is walking on. This information will be important to aid in understanding what the vibro-tactile stimulation might be doing to the perception of the CoP, the movement of the CoP, and the environment itself. Additionally, the distribution of mechanoreceptors is concentrated along the lateral border and forefoot of the plantar surfaces- the same regions the CoP path takes during walking.

With this anatomical perspective, it begins making sense as to how important the foot sole must be in the control of postural and dynamic balance. Further indication of this is the strong connection between these cutaneous receptors and the lower limb muscles. These cutaneous afferents demonstrate strong coupling with spinal motoneurons that act at the ankle. This was demonstrated through measuring electromyography (EMG) signals of the plantar flexors while stimulating the plantar surfaces (Fallon et al. 2005). It was shown that during stimulation, the EMG activity experienced a coupling of similar periodicity ($R^2 = 0.94$) for all the different types of cutaneous mechanoreceptors. This shows how strong of an effect the plantar surface cutaneous receptors have on muscle activity, and thus balance control.

Animal Models

Now with the anatomical knowledge of how these cutaneous receptors are interpreted and strongly connected to motor outputs, it would be important to understand their connection and use for gait itself. Sherrington demonstrated in 1910 that walking gait can be performed without input from the brain, as demonstrated through decerebrated cats (Sherrington 1910). However, this phenomenon is not possible without proper cutaneous feedback from the plantar surfaces of the feet (Slawinska et al. 2012). When performing a similar experiment with rats, it was found that applying a numbing agent, lidocaine, to the hindfeet would stop proper hind-limb coordination and hind-limb fore-limb coordination (Slawinska et al. 2012). Even strong effects to gait cycle duration were seen (Slawinska et al. 2012). Therefore, specifically cutaneous afferents are vital for the control of gait.

Studies with feline models demonstrate the impact these receptors have on the temporal control of gait, the level of foot lift during the swing phase, and importantly allowing for better control of step placements on a step-to-step basis (Rossignol et al. 2006). Cats had five cutaneous nerves in their foot pads severed, then performed a precision task of walking on the rungs of a horizontal ladder. The denervation led to the cats being unable to properly place their feet on the rungs for 3-7 weeks, afterwards they learned to do so by gripping the rungs with a claw-like position (Rossignol et al. 2006). Additionally, having the same cats walk on a tilted treadmill led to them walking in an unstable manner. A corrective measure the cats seemed to take was increasing their step width for enhanced balance through increasing their base of support. This demonstrates one of the primary roles of the cutaneous receptors during gait, giving the CNS information about the walking surfaces, and with such information enhancing the ability for proper foot placements and CoP placement (Rossignol et al. 2006).

Further indication of the importance of plantar cutaneous receptors in proper foot placement and locomoting in sloped terrains comes from a study that used mice to genetically remove a set of interneurons involved with transmitting tactile information to the CNS within the postsynaptic dorsal column of the spinal cord (Paixao et al. 2019). After validating the knocked-out interneurons were involved in the perception of fine touch, gait control was investigated using textured environments and light touches to the feet. No differences were found in walking the patterns of the mice when walking on a level surface, however the mice without the interneurons had significantly more hindlimb slips while traversing a narrow-declined beam (Paixao et al. 2019). Amazingly, knocking out these interneurons did not remove the sense of pain or itching in the affected regions,

because these sensations are carried through different nerve fibers and separate receptors (Paixao et al. 2019). Therefore, these gait control effects are specifically from the fine touch mechanoreceptors (as described in *Cutaneous Receptors*).

These studies give some indication to the use of plantar tactile afferents in the control of gait. Specifically, it seems that they are very important in proper foot placements and step width in complex walking tasks, such as walking a narrow-inclined beam (Paixao et al. 2019) or walking on a tilted surface (Rossignol et al. 2006). Additionally, these animal studies demonstrate that the plantar afferents may not play as vital of a role in balance and gait control during level walking. Therefore, vibro-tactile stimulation effects may not be as strong during level walking as they would during inclined or declined walking where further information about the environment may be needed. This would make sense, due to one suspected role of plantar tactile information is to compare the location of the CoP to the rest of the body (Roll et al. 2002). When traversing a more complex environment, such as inclined walking, there is a higher risk of a slip (McIntosh et al. 2006) thus this relation between the CoP to the CoM has more importance in maintaining balance.

Section 2: Plantar Surfaces of Healthy Subjects

Removal of Plantar Afferents

Like a proper geneticist, before investigating the effects of altering the variable to interest, one approach is to remove the variable and see what happens. In our case, it is important to understand the effects of removing plantar afferents on standing balance. Firstly, with 10 healthy subjects it was found that desensitizing only the forefoot region

with ice did not shift the CoP anteriorly or posteriorly, but instead led to instability in the mediolateral direction while eyes were closed. In contrast, desensitizing the whole foot led to strong effects in the postural control in the anteroposterior direction. However, both desensitizing conditions resulted in increases in CoP velocity (Meyer et al. 2004). The authors attributed these results to represent that the forefoot is predominantly in control of posture in the mediolateral direction, while the heels are involved in the control of posture in the anteroposterior direction.

Concerning what removing plantar afferents does to gait, one study investigated the effects of plantar cooling on pressure distributions and EMG activities during walking. To do this, 10 healthy young subjects walk after cooling specific regions of the feet to a temperature of 6°C while measuring the position of the CoP and muscle activations of the lower limbs (Nurse & Nigg 2001). They found that when a region of the foot was desensitized there was altered muscular activity, and CoP shifts away from the desensitized region to regions that still had normal sensation. This demonstrates: 1) the cutaneous receptors have an immediate effect on the control of gait and the muscle outputs themselves, and 2) the differences in pressure distribution along the foot, shown by the CoP, maybe a major source of information about the environment and how to respond appropriately.

How the removal of plantar afferents could affect balance control would be especially important to understand. First, a brief explanation of the extrapolated center of mass (XCoM) and the margin of stability (MoS) is needed. The XCoM is a measure of where the CoM is during a task and considers where it will end up based on its velocity (Hof et al. 2007). This is done by adding the velocity of the CoM to the position of the

CoM (Hof et al. 2007). It follows then that to keep the CoM within the base of support (BoS), which is required for stable gait, the BoS must be placed lateral to the position of the XCoM (Hof et al. 2007). Therefore, a measure of how close to XCoM gets to this BoS indicates the level of stability the individual has during walking (Hof et al. 2007).

Now, one study attempted to investigate how reducing the plantar cutaneous afferents affected this stability measure while adapting to different stiffness levels of a surface (Hohne et al. 2011). When the plantar surfaces of study participants were anesthetized, they were still able to adapt to the soft surface, however, they did so with a significantly larger MoS than the control group (Hohne et al. 2011). This demonstrates that healthy individuals can adapt to different surface conditions even without plantar cutaneous afferents. Based on how the initial response to stepping on the soft surface resulted in a greater MoS in the plantar numbing group, it is possible that these individuals learned to adapt to the walking conditions using a different method, such as proprioception. Being able to have cutaneous afferents seems to allow for faster and more efficient adaptation to changes in the environment, which can have a direct impact on stability measures such as the MoS.

Studies in clinical populations, such as those with peripheral neuropathy help us understand how the absence of plantar cutaneous afferents affects gait. One source of peripheral neuropathy is from metabolic and microvessel alterations due to hyperglycemia from diabetes (Alam et al. 2017). One prominent symptom of this disease is a loss of sensation of the foot (Alam et al. 2017). Due to this loss of sensation, people with diabetic neuropathy are at a far greater risk of falling than people with plantar sensation (Alam et al. 2017; Cavanagh et al. 1992). Along with this increased risk of

falling, peripheral neuropathy patients also show reduced walking speed, and more variable step lengths and velocities (Alam et al. 2017).

Stimulation During Standing

Vibro-tactile stimulation was found to alter postural control in healthy individuals. In one study, 10 healthy subjects stood barefoot with their eyes closed on a surface with two electromagnetic vibrators (Kavounoudias et al. 1998). The vibrators were separated such that vibrations applied to the forefoot and rearfoot could be independently controlled. It was found that if vibrations were applied to a particular region of the foot, the body leaned away from that region. This was true even if only one foot was stimulated. For example, when the rear of the right foot was stimulated, the individuals would lean forward by shifting their CoP over 20mm forward and about 10mm to the left (Kavounoudias et al. 1998). These results clearly demonstrate that the cutaneous receptors within the plantar surfaces are involved in whole-body balance and postural control. The vibrations were attributed to be simulating increased pressure on that region of the sole, thus causing the CoP and whole-body shifts.

A later experiment investigating how different frequencies may alter this found CoP shift. In this study, nine healthy individuals performed the same task as described previously (Kavounoudias et al. 1998). However, now they tested the effects of two other frequencies, 20 and 60Hz. They also tested the effects of different frequencies to different regions at the same time. For example, one condition involved the forefoot being stimulated with 20Hz while the rearfoot was stimulated at 100Hz (Kavounoudias et al. 1999). It was found that the CoP would shift away from regions that had the vibrations, however, this CoP shift away from stimulation was greater with higher frequencies

(Kavounoudias et al. 1999). The average CoP shift was 15mm for 20Hz, 20mm for 60Hz, and 40mm for 100Hz. These findings were similar when applying different frequencies simultaneously: the greater the difference between these frequencies the greater the shift away from the higher frequency vibration (Kavounoudias et al. 1999). These postural shifts were attributed to the vibrations causing an increase in sensitivity to pressures applied to the stimulated area. The individuals then shifted their CoP away from that region to return to an equal pressure distribution across the plantar surfaces, which would be representative of being vertical.

These results could suggest what vibro-tactile stimulation does to the perception of the environment, through augmenting the feedback from the plantar mechanoreceptors. Based on the effects of vibro-tactile stimulation on postural control, high frequency vibrations may impact the sense of pressure from the mechanoreceptors responsible for the perception of the level of pressure applied to the skin - Merkel cells (Gardner 2010). However, there is no supporting evidence that Merkel cells are specifically sensitive to mechanical vibrations to the skin (Gardner 2010). Mechanical vibrations of 100Hz are within the sensitivity range of Meissner and Pacinian corpuscles (Zimmerman et al. 2014; Gardner 2010). However, these mechanoreceptors on their own don't give indication to a level of pressure applied to the skin (Zimmerman et al. 2014). An alternative reasoning for the postural shifts away from the vibro-tactile stimulation is based on the responses of a lack of tactile sensation (Meyer et al. 2004, Nurse & Nigg 2001). It could be that the body leans away from the vibrations because the high frequency vibrations apply too much noise to the mechanoreceptors in that region. Thus, the body shifts the CoP away

from that region to gain more accurate information about the environment for better postural control.

Stimulation During Stepping

Another test involving high frequency tactile vibrations investigated 20 healthy young individuals stepping onto a sub-threshold vibrating surface that could change in compliance (Visell et al. 2011). It was found that varying vibrations would cause the participants to perceive a higher surface compliance than without the vibrations. This was attributed to the vibrations increasing the perceived displacement during stepping. It is unclear if this increased perception of displacement would be beneficial or detrimental in appropriately responding to a complex walking surface. There are two possibilities: this enhanced perception gives quality information that could lead to more accurate and faster responses to environmental changes, or the enhanced perception is faulty/noisy such that balance responses are negatively affected.

Stimulation During Gait

Supra-threshold tactile stimulation has been shown to alter gait dynamics, specifically at the preferred walking speed (PWS) of healthy individuals. Applying vibrations to 10 healthy young subjects during walking at PWS decreased the long-range correlations of the stride times ($p = 0.014$) when compared to no stimulus (Chien et al. 2017). A decrease in multiscale characteristics of gait variables can be interpreted as an increase of the degrees of freedom for the system, which allows for further flexibility and adaptability to the environment (Jordan et al. 2007). These effects would be very beneficial to chronic stroke survivors.

This type of tactile stimulation was even found to demonstrate the flexibility of the CNS while performing a split-belt task (Mukherjee et al. 2015). The split-belt task causes the two limbs to move in different velocities while walking, a paradigm that is commonly used to show the adaptability and flexibility of the CNS to complete a walking task (Mukherjee et al. 2015). To do this, 10 healthy young subjects performed the split-belt task with vibration stimulation and 10 others performed the task without stimulation. The stimulation had no effect on learning the split-belt task, with both groups adapted to the novel environment equally (Mukherjee et al. 2015). This demonstrates the flexibility of the CNS, even with the presence of stimulation to the plantar surfaces, effectively altering the afferent information, adaption was still able to take place. However, the groups diverged when testing the transfer effect of the split-belt task. For this, they had the participants walk overground, with no stimulation to either group, to see if they transferred the asymmetric walking pattern from the split-belt to over ground. It was shown that the stimulation led to a longer transfer effect, meaning the stimulation group persisted in the asymmetric walking longer than the control group (Mukherjee et al. 2015). This indicated different mechanisms of learning the split-belt adaptation task with and without tactile stimulation. Specifically, the stimulation decreased the reliability of the tactile afferent information from the cutaneous receptors, which consequently increased the reliance/weight of the afferents coming from the proprioceptive system.

While the removal of plantar cutaneous afferents led to changes in the adaptation to different surface stiffnesses (Hohne et al. 2011), applying vibro-tactile stimulation led to changes in adapting to a completely different walking task (Mukherjee et al. 2015). These effects in adapting to the environment demonstrate one of the main purposes of the

feedback from the plantar foot surfaces during gait, and that vibro-tactile stimulation seems to alter this purpose. This provides us the motivation to delve deeper into these observations and determine whether tactile stimulation that stimulates the specific sequence of foot contact during gait also has specific effects on gait patterns.

Visual Conflict

There is an important note to make concerning the effect visual information might have on the interpretation of plantar tactile stimulation. The CNS receives redundant information from all the different sensory systems (Mackinnon 2018). Different factors such as task constraints (walking, running, etc.) may make certain sensory feedback less reliable than others. Therefore, the CNS tends to eliminate unnecessary computational resources by putting a weight on each sensory feedback modality depending on task, environmental or individual constraints and selects an optimal behavioral outcome based on a multisensory integration model (Eikema et al. 2013; Peterka & Loughlin 2004). Our objective is to test the specific nature of tactile feedback information from the foot plantar surface during walking on surfaces of different inclines.

One such way to test how the different sensory systems are weighted is seeing how learning takes place during a split-belt task while removing specific sensory systems. When vision is removed, larger aftereffects of the split-belt task remained when compared to the presence of vision (Torres-Oviedo & Bastian 2010). This demonstrates that without vision, individuals were able to learn the split-belt task better than with the presence of vision. The authors attributed these findings to vision having a larger sensory weight than the somatosensory system during walking (Torres-Oviedo & Bastian 2010), which was detrimental to the learning of the task due to the task being predominantly a

test of the somatosensory system (Hoogkamer 2017). It is therefore possible that the presence of vision during stimulation of the plantar surfaces, may reduce the weight of the somatosensory system and keep it too low to have a discernible effect on gait patterns. This will be further discussed in Section 3.

The ultimate aim of this study is gain insight on the effects of tactile stimulation in stroke survivors. However, first finding what this stimulation effect is on healthy individuals will give further indication to how it may be useful for a rehabilitation tool.

Section 3: Stroke Survivors

This study's ultimate aim is to use a stimulation device towards the population of stroke survivors. There are two main reasons why: the prevalence of stroke in the world, and the specific effect of stroke on the brain. According to the World Health Organization, stroke is the second leading cause of death and third leading cause of disability in the world (WHO, 2021). Therefore, any advancements in rehabilitation for stroke survivors would benefit millions of people. The stroke itself causes a lack of blood flow to a particular region of the brain, which causes cell death to a set of neurons, leading to what is called a lesion (Grefkes & Ward 2014). This means that the rest of the brain must regain any functional role those dead neurons had in daily life. Thankfully, the brain is very adaptable and can perform neuroplastic changes for the surrounding existing neurons to replace the function of the lesioned neurons (Grefkes & Ward 2014). This phenomenon is called neural reorganization and it is vital for the recovery of function after a stroke (Grefkes & Ward 2014). A good way in describing how this reorganization

works is the brain relearning how to perform specific functions that were lost due to the lesion. However, just like learning any new skill, practice and proper feedback is needed.

Concerning balance control, feedback from the peripheral sensory systems is needed to relearn how to properly control locomotion. Unfortunately, it is common for stroke survivors to have sensory deficits in the lower limbs, including the tactile sensitivity on the plantar surfaces (Carey et al. 1993). This is where tactile stimulation could aid in this relearning process. Through adding tactile stimulation to the plantar surfaces during gait, the cutaneous afferent feedback will be more pronounced, bringing attention to those afferents. Based on how important those afferents are in foot placement, step width, and other mechanisms of balance control (as described in *Sections 1 & 2*) the relearning process could thus be accelerated.

Continuing on the effect of vision, in the case of stroke survivors, this visual reliance is exacerbated. There is a prevalence of individuals with post stroke hemiplegia to have immense reliance on their visual information, and difficulty focusing on vestibular and somatosensory inputs even with the absence of visual information (Perennou et al. 2002; Bonan et al. 2004). A behavior that demonstrates this is the 'Pusher' behavior (Perennou et al. 2002). Some stroke survivors tend to have an altered sense of what is vertical with the absence of visual inputs. This altered internal sense of what is vertical is enough for the individual to begin pushing against someone that might be trying to keep them vertical according to their actual environment, while to the Pusher the other person is in the way of being vertical in space (Perennou et al. 2002). However, once visual information returns, the stroke survivor can see that their internal sense of vertical is not the actual external vertical and thus correct their upright posture (Perennou

et al. 2002). This demonstrates that many stroke survivors focus predominantly on their visual information with low to negligible reliance/weightage for the other sensory modalities.

Due to this increased visual reliance, being able to train stroke survivors to rely on their other senses could increase their balance control (Bonan et al. 2004). A total of 20 stroke survivors took part in a 4-week balance rehabilitation program, half with vision and the other with the absence of vision during the rehabilitation. Balance tests were then performed on the stroke survivors before and after the program using a sensory organization test (SOT). It was found that all stroke survivors increased their balance control, however, the group that performed the rehabilitation without vision scored better than those who performed the rehab with vision (Bonan et al. 2004). The authors concluded that after a stroke reliance on visual information increases, possibly through a decrease in the somatosensory and vestibular sensitivity, and this negatively affects balance control due to balance being a multi-sensory task. If this balance training can increase the effectiveness and trust in the vestibular system of the stroke survivors, maybe something similar can be done for the somatosensory system through increasing the level of stimulation.

Stimulation During Walking – Stroke Survivors

Vibro-tactile stimulation has been used in other locations besides the plantar surfaces. One such study used it as a biofeedback tool to decrease stride length and time asymmetry in chronic stroke survivors (Afzal et al. 2015). This was done by applying vibrations to the calf of the paretic limb for the same amount of time of the previous non-paretic step, or by having vibration intensity decrease as asymmetry decreased (Afzal et

al. 2015). Both stimulation methods increased symmetry between the paretic and non-paretic limbs when compared to the control no stimulation condition (Afzal et al. 2015). An argument could be made that if the stroke survivors are unable to properly perceive or interpret the tactile afferents from the plantar surfaces then applying vibration to another region of the body as a feedback mechanism could be more beneficial as a rehabilitation tool. However, this would require the stroke survivors to learn what the stimulation type represents in this new region and understand how to use such information in their balance control. This could take many training sessions and require a lot of mental effort from the stroke survivors. Additionally, if they were able to learn to use the device properly it could still lead to a dependence on such a device. If instead, the stimulation was applied to the plantar surfaces, neuroplastic changes could take place that could help recalibrate the sensorimotor apparatus with the correct stimulus from the plantar surfaces. With such neuroplastic changes, there would be a decrease in dependency on the device.

Concerning studies that have investigated the effects of stimulating the plantar surfaces of chronic stroke survivors, a majority have investigated the effects of sub-threshold stochastic resonance vibrations on balance (see review: White et al. 2018). The reasoning behind the stochastic resonance vibrations is that it has a resonance effect on sensory thresholds and reduces postural sway. However, such sway reduction may be detrimental to balance control during gait. Sub-sensory vibrations do not augment the tactile information, it would only lead to a slight increase in sensitivity. While adding above threshold vibrations has not been investigated as thoroughly and could lead to augmenting the tactile information to enhance attention to the plantar surfaces and aid in neuroplastic changes.

There are a limited number of studies that have investigated the effects of supra-threshold vibro-tactile stimulation to the plantar surfaces of chronic stroke survivors. One study tested the effects of such vibro-tactile stimulation during treadmill walking at a self-selected speed (Liang et al. 2021). The stimulation was given to the plantar surfaces throughout the entire trial. They found little to no effects to kinetic or kinematic variables of the stroke survivors (Liang et al. 2021). This lack of effect from the stimulation could be from the stimulation not being used to give additional attention/information about the environment the subjects were locomoting in. Specifically, such stimulation should intuitively follow the natural patterns of plantar mechanoreceptor feedback as it occurs during walking. If the stimulation were to be given only during the stance phase of gait, as well as follow the path of the CoP much larger positive effects may be elicited.

Section 4: Brain Imaging

First, the CNS seems to have a particular sensitivity to the speed at which stimuli move across glabrous skin, such as the palm of the hand (Oh et al. 2017). When testing different velocities of stimuli moving across the palm of the hand on brain responses, it was found that an intermediate speed had the greatest brain response compared to a faster and slower speed (Oh et al. 2017). Specifically, 65cm/s was found to have the least amount of BOLD responses, then 5cm/s, and 25cm/s had the greatest level of BOLD responses (Oh et al. 2007). The reason for these speed-dependent responses is left up to speculation, it could be that being more responsive to speeds close to 5cm/s and 25cm/s is beneficial in the manipulation of objects. It would be interesting to test if this same speed-dependent responses are present for plantar stimulation as well. Furthermore, it could be that the brain would be most sensitive to the speed at which the CoP traverses

across the foot during normal walking. Based on the importance of this CoP in balance control, as previously described in Sections 1&2, the brain could be actively paying more attention to this CoP movement and speed to have proper gait control.

Describing how plantar tactile information is processed in the brain would aid in the understanding of how the tactile information may be used in the control of balance and gait. Some studies have used MRI-compatible foot-sole stimulators to apply walking related tactile stimuli in order to see where in the brain such information is processed. One such device takes pressure data from a walking trial and mimics the pressure distribution on the foot using air cylinders and a movable plate within a boot (Zhang et al. 2019). All of this is performed while the subject is supine in the MRI. With such a device, a better representation of what brain areas are involved in the perception and sensory motor integration of tactile information from the plantar surfaces can be done. Significant brain activity was found in the supplementary motor area (SMA), primary motor cortex (M1), inferior parietal lobule, middle temporal gyrus, and hippocampus (Zhang et al. 2019). It is important to note that by simply applying pressures to the plantar surfaces, specific regions of the brain are activated. Specifically, the SMA which is important for voluntary motor tasks (Burton et al. 1996), as well as correcting posture during motor tasks (Brinkman 1984; Nakagawa et al. 2016) and M1, which is associated with the actual control of individual limbs for movements (Sanes 2000).

To further drive this point, another MRI compatible plantar pressure stimulator was used to find the differences between a gait-like organized and a random plantar stimulation pattern. Additionally pneumatic stimulation below the foot was compared to mental imagery of walking (Labriffe et al. 2017). Although the two stimulation patterns

did not show any differences in brain activation, the stimulation and mental imagery conditions showed similarity for activity at the SMA and differences at M1 with only the tactile stimulation activating the motor cortex. The similarity between the two tactile stimulation conditions was attributed to the different patterns not being different enough. One feature of the Korvit boots that might have caused this was how each boot could only stimulate the forefoot and/or the rearfoot. Therefore, stimulating the plantar surfaces in more places and in complete opposite patterns could lead to more distinctive changes in afferent information. Due to both imagining walking and simply stimulating the plantar surfaces activating similar brain regions, it would be reasonable to assume this plantar tactile information is used by the CNS for online adjustments in gait control.

Finally, how does a stroke affect the perception of cutaneous stimulation? Due to various places a lesion from a stroke could occur, as well as the size of the lesion, there is not one common symptom of a stroke. It could lead to a complete absence, decrease, increase, or distortion compared to normal sensory sensation (Lv et al. 2022). For example, the thalamus is involved in sending sensory afferents to the cortex and modulating cortical processing, thus if a lesion occurs there it can cause deafferentation and a loss of somatosensory information entirely (Lv et al. 2022). However, if a lesion occurs in the somatosensory areas of the parietal cortex, then only one specific region on the body may have somatosensory deficits (Lv et al. 2022). To better understand how the area of the lesion may alter perception and motor output an analysis called Voxel-Based Lesion-Symptom Mapping (VLSM) has been developed (Lv et al. 2022). This analysis calculates the statistical contribution of damaged voxels (Three-dimensional measuring units of MRI) to a certain sensation symptom through normalization of multiple stroke

survivor MRI results (Lv et al. 2022). There are currently very few, if any, studies investigating how a lesion alters the whole brain activity in stroke survivors. Therefore, this study would add to the literature to aid in this VLSM analysis. This in turn would aid in the understanding of how a lesion in the brain could alter this important perception and use of the tactile afferents coming from the plantar surfaces for the control of gait in chronic stroke survivors.

Section 5: Summary

There are many studies that show how important of a role the plantar cutaneous receptors play in gait. Animal studies show that it is important for foot placement, completing complex gait tasks, and walking on sloped surfaces (Rossignol et al. 2006, Paixao et al. 2019). They aid in these tasks through giving information about the environment that is being traversed (Meyer et al. 2004, Hohne et al. 2011, Visell et al. 2011). One way the plantar surfaces seem to gain such information is through the location of the CoP (Meyer et al. 2004). However, during gait this CoP traverses along the foot; thus, the foot must have the ability to locate and interpret this moving stimulus.

Anatomically, the foot has the resources to do so. Fast adapting fibers and mechanoreceptors that are sensitive to a moving stimulus make up the majority of the touch receptors within the plantar surfaces (Stzalkowski et al. 2018, Zimmerman et al. 2014). These mechanoreceptors and fast adapting fibers seem to be organized along the foot to pick up the path of this CoP during walking (Stzalkowski et al. 2018, Corniani & Saal. 2020). These same mechanoreceptors are also not only sensitive to moving stimuli, but also vibrations or different frequencies (Zimmerman et al. 2014). One of these

mechanoreceptors, Meissner's Corpuscles, are thought to be involved in detecting when a slip occurs and are sensitive to low frequency vibrations (Zimmerman et al. 2014).

When applying vibrations to the plantar surfaces during quiet stance, individuals shift their CoP away from the vibrations (Kavounoudias et al. 1998). This CoP shift is even frequency dependent. Individuals will shift their CoP further away from higher frequency vibrations than lower frequency vibrations (Kavounoudias et al. 1999). This could suggest either a change in the amount of perceived pressure in the high frequency vibrated regions (Kavounoudias et al. 1998 & 1999), or a decrease in proper tactile feedback (Meyer et al. 2004, Nurse & Nigg 2001). Lower frequency vibrations could augment such feedback to the CNS.

However, the need for this tactile information changes based on the walking task (Rossignol et al. 2006, Torres-Oviedo & Bastian 2010). Vibro-tactile stimulation might be able to alter the sensory weight of plantar tactile information when learning a new task (Mukherjee et al. 2015). One particular task that could increase the need for this tactile feedback is walking on an incline (Rossignol et al. 2006, Paixao et al. 2019, Sun et al. 1996). One such reason could be that inclined walking leads to increases shear forces with the walking surface, and thus increase the risk of a slip (Sun et al. 1996). With mechanoreceptors in the plantar surfaces being responsible for alerting the CNS of a slip and sensitive to low frequency vibro-tactile stimulation, adding such stimulation during inclined walking could increase the effect of such stimulation on gait when compared to level ground.

Applying vibro-tactile stimulation in different patterns to plantar surfaces during gait would then give indication to how strictly the afferent information from the plantar

surfaces can be augmented to affect gait. This could further elaborate on how this afferent information is processed within the CNS.

Plantar tactile afferents are strongly linked to brain areas that are also involved in movement control (M1) as well as movement planning and adjustments (SMA) (Zhang et al. 2019, Labriffe et al. 2017). Dynamic tactile stimuli have shown the brain to be sensitive to specific speeds of tactile stimuli (Oh et al. 2007). This was shown on the hand to have a heightened sensitivity to 25cm/s. It could be that for the foot this speed sensitivity has been habituated to be the speed the CoP moves across the foot during normal walking. Therefore, stimulating the plantar surfaces in a manner that follows the CoP at the speed at which the individual walks most often, their preferred walking speed, may alter how the brain responds to that CoP movement.

A stroke can negatively affect how this plantar tactile feedback is used. Stroke survivors can have decreased tactile sensitivity in their lower limbs (Carey et al. 1993). However, this is not from peripheral problems in how the afferent information is carried to the CNS, like how peripheral neuropathy patients are affected (Alam et al. 2017). Instead, it is from a loss of a set of neurons that were involved in multiple tasks (Grefkes & Ward 2014). Thus, the individual must relearn how to use such feedback to perform a task (Grefkes & Ward 2014). With redundant sensory information coming to the CNS from all sensory systems, the individual may choose to focus on one particular sensory feedback system to control movements. With stroke survivors having high reliance on visual feedback during balance tasks (Perennou et al. 2002; Bonan et al. 2004) they may choose to only focus on visual information when using tactile information. Thus, applying supra-threshold stimulation to the plantar surfaces, increased attention and

reliance on the tactile information could occur. Specifically, stimulating in a manner that emphasizes the tactile stimuli of importance, the CoP, could aid in the recovery of balance control more than applying vibro-tactile stimulation across the whole foot at once (Liang et al. 2021), and stimulating in a pattern that does not follow this CoP movement could instead negatively impact balance control through an even further faulty perception.

Therefore, the following chapters discuss the development and validation of a real-time vibro-tactile stimulation system that can be used during gait, and implementing this system to see the effects on gait and balance control in healthy individuals. This stimulation system can provide vibrations to the plantar surfaces in different sequences, one that follows the natural progression of the CoP during walking, as well as an unpredictable abnormal sequence. We then implemented this system in an investigation of how these different sequences alter healthy adult walking at different inclines and with low visual information. This was done to better understand how the plantar surfaces are involved in gait and balance control.

Chapter 3: Development of Real-Time Augmented Vibro-Tactile

Stimulation During Gait

Introduction

The plantar surfaces are the only part of the body that are in direct contact with the environment during independent walking. It is known that the plantar surfaces are important for balance control during walking through the feedback they supply about the environment (Inglis et al. 2002). This is done through biological touch sensors, called mechanoreceptors, within the skin (Meyer et al. 2004). The removal of this sensory feedback has been shown to cause negative effects during postural control (Meyer et al. 2004).

For this reason, there have been many studies investigating methods of adding additional stimulation to the plantar surfaces with the objective of helping clinical populations that suffer from reduced plantar sensitivity (Xie et al. 2023). One such method is supplying vibrations to the plantar surfaces during walking (Chien et al. 2017, Mukherjee et al. 2015, Liang et al. 2022). However, many of these studies supply such vibrations independent of the phases of the gait cycle. These vibrations are instead typically supplied throughout an entire gait cycle, including the swing phase (Xie et al. 2023). This would therefore be perceived as a foreign stimulus to the plantar surfaces, since the feet are not in contact with the ground. In fact, when measuring afferent neuronal activations from mechanoreceptors in the foot, no passive neuronal activations were recorded when the foot was in the air (Kennedy et al. 2002). Thus, supplying stimulation to the plantar surfaces during swing would instead be a novel perturbing

stimulus, as opposed to aiding in the perception of the environment during natural foot contact.

Therefore, creating an online vibro-tactile stimulation system that only stimulates the foot during the stance phase of gait would allow for a more accurate interpretation of the effects of augmented plantar feedback on gait and balance control during walking. Only a few studies have linked plantar vibrations to the stance phases of the individual during walking (Novak & Novak 2006, Toda et al. 2020, Toda et al. 2022). Toda and colleagues vibrated the nail of the hallux during the stance phase of walking (Toda et al. 2020, Toda et al. 2022). While Novak & Novak had plantar vibrations activate from pressure sensors within the insoles of the shoes, leading to vibrations going from the heel to the forefoot- following the natural progression of stance (Novak & Novak 2006). These studies had the stimulation tied with the stance phase, with one even matching the progression of stance at two sites (Novak & Novak 2006). However, the majority of plantar stimulation studies have at least three sites of stimulation: the heel and under the base of metatarsal (MT) joints 1 and 5 (Chien et al. 2017, Mukherjee et al. 2015, Liang et al. 2022, Novak & Novak 2006, Galica et al. 2009, Pathak et al. 2022, Song et al. 2012, Stephen et al. 2012). Having the activation of these three sites be sequentially activated would allow for precise testing on how the plantar surface activation may supply information gained about the environment to the brain.

For these reasons, we created an online vibro-tactile stimulation system that takes live marker position data from the subject to activate individual tactor sets timed according to the stance phases of gait. Additionally, the system can either activate tactors sequentially to follow the normal progression of the stance phase or activate tactors in an

unnatural sequential or random pattern. In this paper, we describe the developmental process of our online vibro-tactile stimulation system where we describe the equipment setup, the software algorithm used to detect gait phases, how activation signals were sent to the tactors, testing dampening effects during walking, enhancing tactor signals with use of washers, designing the sequential tactile stimulation and finally testing and validating the presence of the applied tactile frequency during walking.

Methods

Equipment

A total of 12 C-2 tactors (Engineering Acoustics Inc., EAI, Casselberry, FL), six within each insole, were used for supplying the vibro-tactile stimulation. These tactors were set to vibrate at a frequency of 250Hz, and an amplitude of 23.5db (2.1Vrms). This frequency was chosen to mimic the vibrations used in previous studies (Chien et al. 2017, Mukherjee et al. 2015, Xie et al. 2023). Each foot had one controlling box to control the six tactors in the respective insole. These controlling boxes were attached to a fanny pack that the participant wore around their waist (**Figure 1**). Live marker position data was collected by Vicon Nexus (Vicon Oxford, UK) at a frequency of 100Hz and ground reaction force data was collected by an instrumented treadmill (Bertec Version 2.0 2013, Columbus, OH) at 1000Hz.

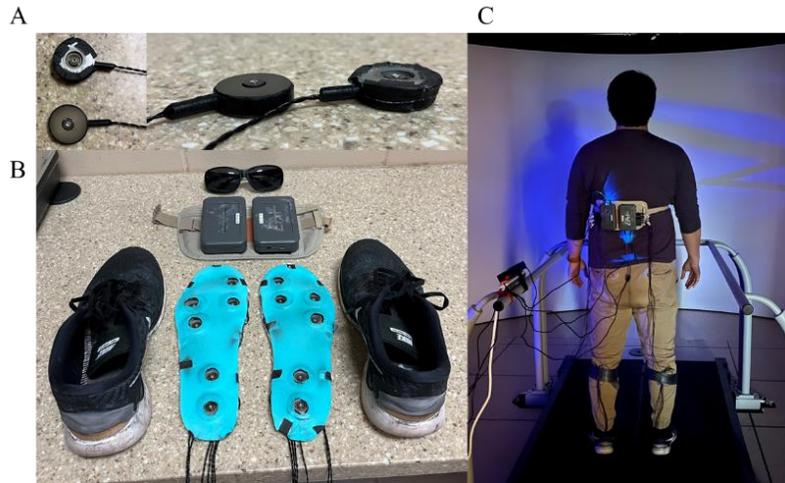


Figure 1: (A) C-2 Tactors had a washer attached to the top to decrease dampening (see Tactor Vibration Damping Testing). (B) Tactor control boxes were attached to a small fanny pack with Velcro and tape. Tactors were embedded within custom-made insoles that fit within the shoes. (C) The wires of the tactors came out of the back of the shoes, then attached to the shank of the subject to limit the wires hitting the legs during walking. These wires were then ordered into the respective side's controller box. The tactor control boxes were supplied power with an extension cord, and were connected to the computer with USB extension cords.

With this data, we first needed a software that could both receive this motion data online and send activation signals to tactors. For this reason, we created the control software in MATLAB (Mathworks, Natick, MA), because Vicon has a DataStreamSDK that can stream collected data of each frame online to MATLAB and EAI supplies a Tactor Control SDK that is compatible with MATLAB (**Figure 2A**).

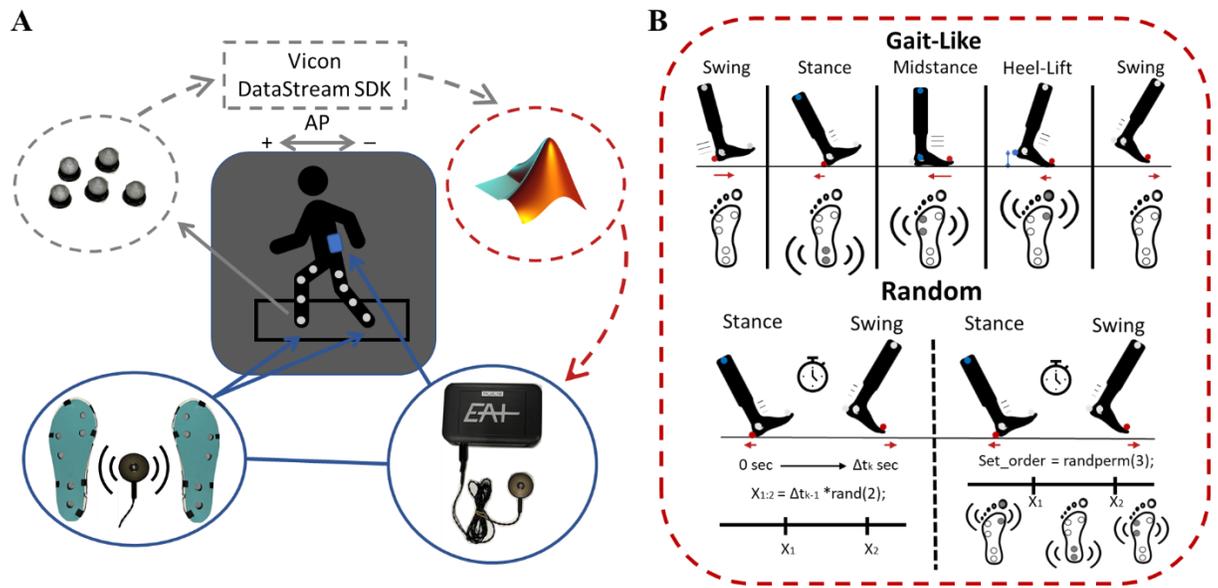


Figure 2: Online work flow for control of tactor embedded insoles through Vicon marker data collected at 100Hz. (A) Within one frame of marker data, the marker positions of the lower body were streamed to MATLAB at 100Hz through Vicon’s DataStreamSDK to activate specific tactor sets during the stance phases of gait. (B) MATLAB used this data to determine when a heel-strike and toe-off had occurred through the velocities of the heel and toe markers respectively. After a heel-strike had occurred, it then either activated different tactor sets based on the relative positions of specific lower body markers or activated randomly based on the time of the previous recorded step. Activation signals were then sent to EAI’s tactor control box to vibrate the specified tactor sets at 250Hz for 30ms. This process was then repeated for the subsequent marker data frames.

Gait Phase Detection

The Vicon DataStreamSDK allows for MATLAB to call for specific data from the current frame. Thus, each collected frame MATLAB obtains the heel and toe marker locations to determine heel strike and toe off. Consequently, when the limb is determined to be in stance, the ankle, hip, heel, and toe markers are used to determine where in the stance phase the subject currently is in (**Figure 2B**).

The moment of heel strike was determined by finding the time point of the heel marker velocity changing from positive to negative. Toe off time point was set as the moment when the toe marker velocity changed from negative to positive (Zeni et al. 2008). These calculations were performed by comparing the heel/toe positions of the current frame with the heel/toe positions of the previous frame. If the velocity changed then stance was set to 0 for swing or 1 for stance (**Figure 3**). Additionally, to control for possible noise or marker position jitter, the factors were only activated or deactivated if this velocity change was maintained for 2 consecutive frames (0.02secs). This process is done for both the left and right feet at the same time.

```

1 - tstart = tic;
2 - while toc(tstart) < Trial_Time
3 -     %% Heel Strike Detection
4 -     % Get current heel position
5 -     L_heel_current = DataStreamSDK(L_heel);
6 -     % Negative Heel Velocity, Previously in Swing, and Velocity has been maintained for 2 frames
7 -     if L_heel_previous - L_heel_current < 0 && L_Stance == 0 && L_Check_HS == 2
8 -         L_Stance = 1; % Stance On
9 -         L_Stance_timer = tic; % Timing Stance Time (for RS)
10 -        L_Check_HS = 0; % Check reset
11 -        % Check that Heel Velocity change is maintained when previously in swing
12 -        elseif L_heel_previous - L_heel_current < 0 && L_Stance == 0
13 -            L_Check_HS = L_Check_HS + 1;
14 -        % In Stance already, or are in Swing
15 -        else
16 -            L_Check_HS = 0;
17 -            L_heel_previous = L_heel_current;
18 -        end
19 -     %% Toe Off Detection
20 -     L_toe_current = DataStreamSDK(L_toe);
21 -     % Positive Toe Velocity, Previously in Stance, and Velocity has been maintained for 2 frames
22 -     if L_toe_previous - L_toe_current > 0 && L_Stance == 1 && L_Check_TO == 2
23 -         L_Stance = 0; % Stance OFF
24 -         L_Prev_Stance_Time = toc(L_Stance_timer) - 2/DataStreamHz; % recording stance time (For RS)
25 -         L_Check_TO = 0; % Check reset
26 -         % Check that Toe Velocity change is maintained when previously in stance
27 -         elseif L_toe_previous - L_toe_current > 0 && L_Stance == 1
28 -             L_Check_TO = L_Check_TO + 1;
29 -         % In Swing already, or are in Stance
30 -         else
31 -             L_Check_TO = 0;
32 -             L_toe_previous = L_toe_current;
33 -         end
34 -     end

```

Figure 3: Example code of how heel strike and toe off was detected. At a rate of 100Hz, the current position of the heel marker ($L_heel_current$) is compared to the last recorded position ($L_heel_previous$). If the current position is behind the previous position, the velocity became negative, and the limb was previous in swing ($L_Stance = 0$), and this negative velocity was maintained for two frames (L_Check_HS

= 2), then a heel strike has occurred ($L_Stance = 1$). Once L_Stance becomes 1 a timer starts to record the current stance time and for progressing to the next random factor set (Figure 4). L_Check_HS is only updated if the heel velocity changed to negative while the limb was in swing. $L_heel_previous$ is replaced at every frame unless L_Check_HS was updated. Toe off is the same process but opposite velocity change during $L_Stance = 1$.

Tactor Activation and Pattern Control

Once the limb is in the stance phase, there are two patterns that were performed: Gait-Like (GS) and Random stimulations (RS). The GS follows the normal progression of stance, while the RS switches between tactor sets at random intervals. For GS, once heel strike is detected, the heel set of tactors activate until midstance is reached. Midstance is detected when the ankle marker reaches the same anteroposterior (AP) position as the hip (the mid-point between the ipsilateral ASIS and PSIS). At this point the tactor sets on the lateral side of the forefoot (MT5) activate until heel lift is detected. Heel lift is determined as the moment when the heel marker rises 3cm above the toe marker and at this point the final tactor set on the forefoot and toes (MT1) activate until toe-off (**Figure 4A**) is detected.

We wanted RS to not just be random tactor sets activated at the same time points as detected from the gait events, but we also wanted them to activate for random durations during stance as well. To do this, we recorded the stance time of the same limb's previous step (Δt_{k-1}). This time was then split into three random sections (X_1) by finding two random percentages (r) of the previous step time (**Equation 1**). When the time after the next heel strike reached this first time point, the first random tactor set

switched to the second set. Finally, when the current stance time reached the next time point, the third factor set activated until toe off (**Figure 4B**).

$$\text{Equation 1: } X_1 = \Delta t_{k-1} * r$$

A

```

36 %% Factor Activations (Gait-Like)
37 % Only Starts if foot is known as being in stance
38 if L_Stance == 1
39     % Before Midstance
40     if L_Ankle_AP > L_Hip_AP
41         % Activate Left foot Factor set 1 (heel) for 30ms
42         Tac_Activate(Left_box, 30, 1);
43     % After Midstance
44     else
45         % Before Heel-Lift of 3cm above Toe
46         if ( L_Heel_Vert - L_Toe_Vert ) < 30
47             % Activate Left foot Factor set 2 (MT5) for 30ms
48             Tac_Activation(Left_box, 30, 2);
49         % After Heel-Lift
50         else
51             % Activate Left foot Factor set 3 (MT1) for 30ms
52             Tac_Activation(Left_box, 30, 3);
53         end
54     end
55 end
56

```

B

```

58 %% Factor Activations (Random)
59 % Creating Factor Durations and Order During Swing for next Stance
60 if L_Stance == 0
61     % Creating Random Factor Activation Durations
62     L_tac_times = sort((L_Prev_Stance_Time).*rand(2,1));
63     % Randomizing Factor Set Order
64     L_Random_Tac = randperm(3);
65 end
66 % Activating Factors During Stance
67 if L_Stance == 1
68     % Before first random time point of current stance phase
69     if toc(L_Stance_timer) < L_tac_times(1)
70         % Activate Left foot Random Factor Set 1 for 30ms
71         Tac_Activation(Left_box, 30, L_Random_Tac(1));
72     % After first but before second random time point
73     elseif toc(L_Stance_timer) < L_tac_times(2)
74         % Activate Left foot Random Factor Set 2 for 30ms
75         Tac_Activation(Left_box, 30, L_Random_Tac(2));
76     % After both random time points (will continue until toe off)
77     else
78         % Activate Left foot Random Factor Set 3 for 30ms
79         Tac_Activation(Left_box, 30, L_Random_Tac(3));
80     end
81 end
82

```

Figure 4: (A) Example code of how factors were activated in a gait-like sequence. Once the limb entered a stance phase the heel set was continuously sent to activate for 30ms, resulting in continuous vibrations. Then when the ankle AP position progressed past the hip, the MT5 set was activated. Finally, when the heel lifted 3cm over the toe marker the last factor set at MT1 was active until a toe off. (B) Code showing how factors were activated in a random sequence. During the swing phase of gait, random time points were calculated from the time of the previous step. This allowed for the randomly order factor sets to activate for random durations within the time of the stance phase. Once the limb entered the stance phase, the first factor set activated until the first time point was reached, based on the timer of the current stance phase (L_Stance_timer). Then the subsequent random factor set activated until the second time point was reached. Finally, this last factor set activated until the toe off.

Tactor Vibration Damping Tests

The factors used in this study vibrated mechanically with a linear actuator that moved through a magnetic design (eaiinfo.com/product/c2/); due to the tactors being under the feet while walking, we wanted to make sure the pressures applied would not dampen the mechanical vibration intensity and or frequency. Previous work had tested

vibration perception during sitting, one-leg and two leg standing (Chien et al., 2017) however it was not clear if damping would be a significant issue during walking. Based on preliminary testing, participants mentioned that when pressure was applied to the tactor during walking, they perceived a reduction in vibration intensity. Therefore, we attempted to mitigate this damping by attaching a thin washer (1x3cm diameter with 0.15cm thickness) to the top of the C-2 tactors, then tested if this decreased such damping.

This was done by placing three tactors face down within two foam pads, meant to mimic the skin of the plantar surfaces (**Figures 5A & B**). This was then placed on the force plate along with a 20kg weight on the top piece of padding to supply pressure (**Figure 5B**). The tactors were then activated in 2.5sec pulses with a 0.5sec pause three times to vibrate at 250Hz while vertical ground reaction force (GRF) data was collected. This test was repeated on tactors with and without the attached washers. The addition of the washer led to a 21.18dB increase (almost 300% increase) in the power of the 250Hz frequency component of the vertical GRF data (**Figure 5E**). For this reason, future tests were performed with the washer applied tactors.

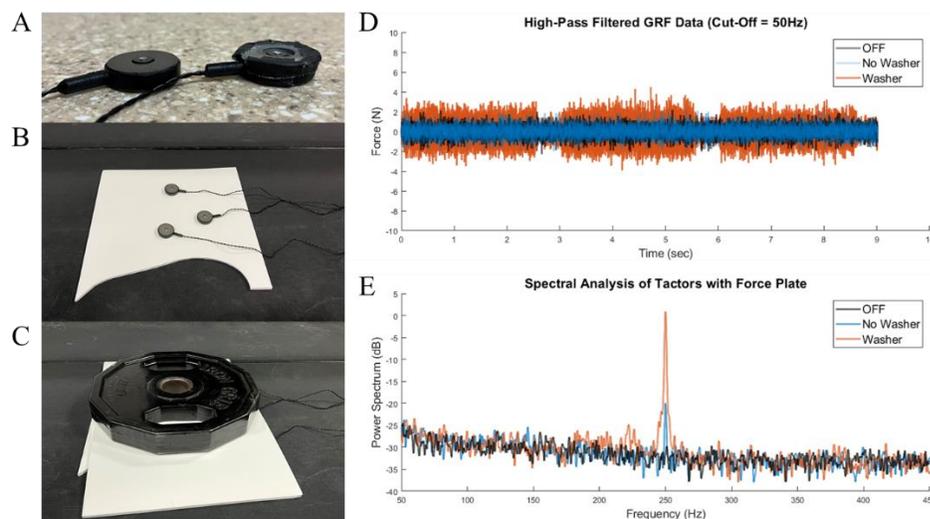


Figure 5: Adding a thin washer to the C-2 tactors allowed for increased vibration perception under pressure. (A) A washer was attached to the C-2 tactors to decrease the dampening of vibrations under pressure. (B) Three tactors were placed face down on a foam pad, (C) then a 20kg weight was placed on top of the tactors with an additional layer of foam to distribute the weight. The tactors were then set to vibrate at 250Hz for three 2.5sec pulses with 0.5sec gaps while vertical GRF data was collected. (D) High pass filtered vertical GRF data with a 50Hz cut-off frequency of tactors OFF, without, and with washers. (E) Spectral analysis of D. Both tactors with and without washers had a 250Hz peak, however, with the added washer, the power of this frequency component was over 20dB larger.

Next, we checked if the changing of pressures applied by the feet would dampen the frequency of the vibrations. An individual walked as the tactor embedded insoles vibrated at 250Hz. However, to be confident that the 250Hz component of the data was not caused by the treadmill moving along the force plate or from the person stepping on the force plates, this test was repeated at three different walking speeds. This was done due to the frequency components of the treadmill and subject would either increase or decrease dependent on the walking speed, whereas the 250Hz vibration would be independent of the movement of the treadmill and subject. At each walking speed the

250Hz frequency component from the tactors remained constant (**Figure 6D**), thus we were confident that the subjects would feel a constant 250Hz vibration during the entire stance phase of each step with the addition of the washer.

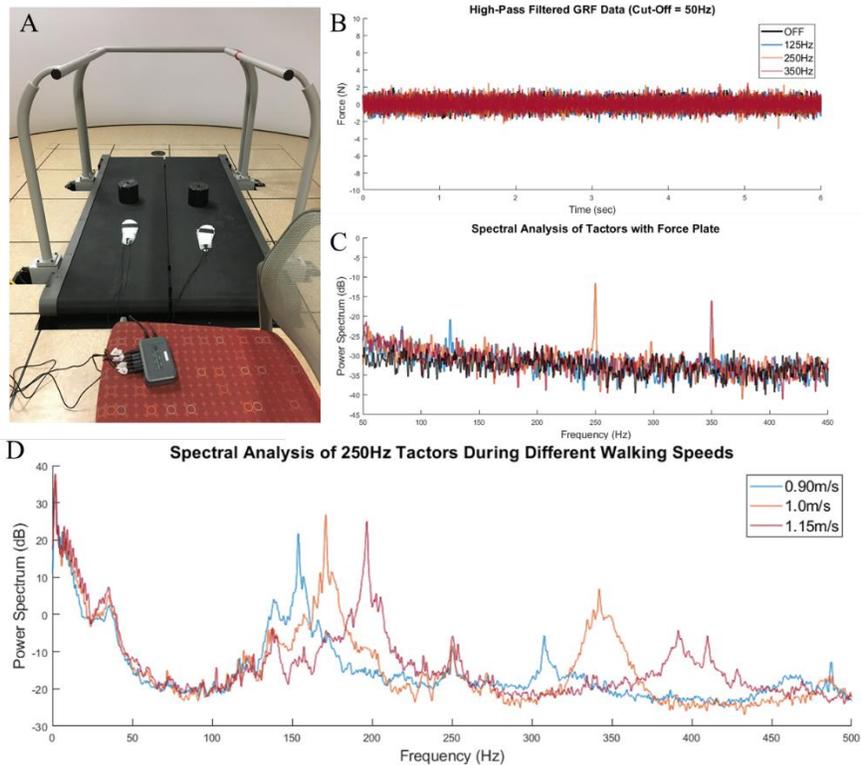


Figure 6: Testing if pressures from walking would alter the frequency of the tactor vibrations. **(A)** Vertical GRF was collected at 1000Hz of stationary insoles sitting on instrumented treadmill, while tactors were either OFF, or set to 125, 250, and 350Hz. **(B)** High-Pass filtered GRF data with a cut-off frequency of 50Hz. **(C)** Spectral analysis of **B** demonstrating a strong frequency component at each respective frequency. **(D)** Spectral analysis of vertical GRF data during walking trials at various walking speeds while 250Hz vibrations were applied to the plantar surfaces. Frequency peaks caused by noise from treadmill or movements of the walking individual increased in frequency as walking speed increased, while 250Hz peak from tactors remained constant at all walking speeds.

Results

Activation Output Timings

From the MATLAB code we were able to have an output of which factors were being activated during each recorded frame. This was then used to find how effective the heel strike and toe off detection method from the code was when compared to measuring from the force plates - the gold standard gait detection method (Zeni et al. 2008). After a walking trial, the heel strike and toe off timings were determined by the vertical GRF passing above a threshold of 20N and sustaining this force for 40ms (Zeni et al. 2008), and the opposite (passing below 40N) for toe off. It was found that the activation and deactivation signals were, at maximum, only 0.04sec delayed when compared to the GRF gait detection method (**Figure 7**). This time delay is similar to what was found by Zeni and colleagues. Additionally, during stance, the three factor sets sequentially activated one after the other going from heel to toe for the GS. The factor sets were activated in a random order and for random durations between subsequent steps for the RS (**Figure 7**).

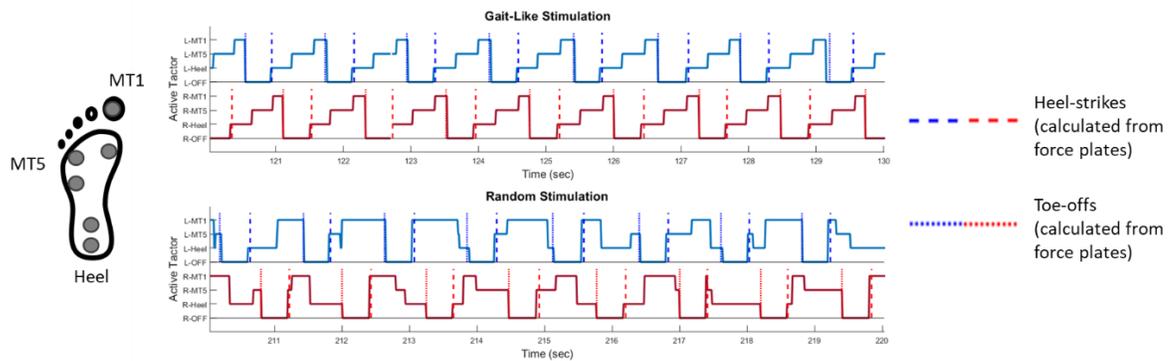


Figure 7: Validation of real-time factor control output from MATLAB to EAI factor control box for GS and RS patterns during level walking. Each step on the y-axis represents which factor set is active at that point of time. Vertical dashed lines represent actual right and left foot heel-strike and toe-off timings when measured by vertical GRF data. Activation signals sent to factors were within 0.04sec of actual heel-strike and toe-off timings. During a stance phase, GS led to a repeated pattern of factor sets activating from heel

to toe, while RS had a random sequence of factor sets activate for random durations for each stance phase independently between the two feet.

Tactor Activations During Each Stimulation Pattern

Next, we performed a single walking trial that consisted of one minute of NS, GS, and RS. This trial was then split into their individual minute-long sections and a spectral analysis of the vertical GRF was done. For GS and RS, a 250Hz peak was present that was not present during the NS section of the data. When subtracting the NS spectral result from the GS and RS sections, it was found that the 250Hz frequency component was over 10dB more powerful than in the NS section (**Figure 8**).

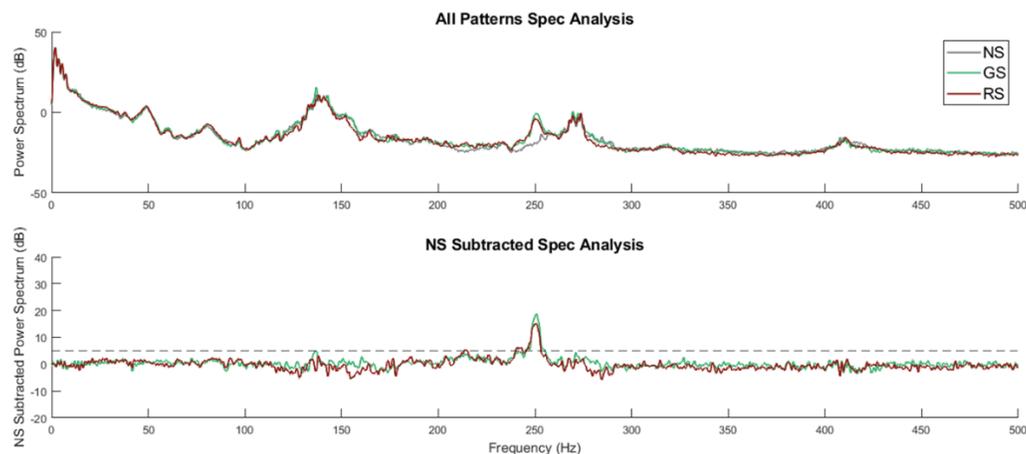


Figure 8: Vertical GRF spectral analysis during one walking trial of both stimulation patterns. (**Top**) The figure shows a total of 3mins of walking data at 0.8m/s split into one-minute durations during which GS (Green), RS (Red), and NS (Grey) were provided. A peak at 250Hz is present for both stimulation patterns that is not present when NS occurred. (**Bottom**) The difference between the two stimulation patterns and the NS to confirm that the 250Hz peak is only present when the tactors were being sent activation signals.

Discussion

In this study, we attempted to create a real-time gait-synced vibro-tactile stimulation system for future studies investigating the effects of plantar stimulation during gait. Based on our validations of the stimulation timings and consistencies, we believe we have succeeded in our purpose. This device is reliably able to stimulate the plantar surfaces in various patterns that follow or go against normal walking behavior.

Additional findings from this study have shown that when using C-2 tactors from Engineering Acoustics Inc. to supply plantar vibrations, the addition of a spacer, such as a washer, can decrease the effect of pressures damping the vibration intensity. Furthermore, these tactors seem to be consistent in the set vibration frequency even when dynamic pressures from walking are applied. It is therefore suggested for future studies when using such tactors in similar studies to use such a method to keep vibration intensity as consistent as possible.

Future studies, when available, should use vertical GRF data to validate that activation signals sent from the controlling unit were received and led to actual tactor activations. In the current study and other collections using this method, there have been many cases for one or both tactor control boxes to disconnect caused by the motion of walking (see **Supplementary Figure I**). The motion of walking led to the wired connection from the computer to the tactor box to be pulled enough to disconnect, but not enough to completely remove the wire from the box. This issue would cause problems especially for studies that perform sub-threshold vibrations, where the subject cannot feel the vibration applied to them (Song et al. 2022). Even during supra-threshold vibration studies, asking a subject if they can feel the vibrations may distract or induce bias on the

results. Therefore, using a Bluetooth connection and or performing a spectral analysis of vertical GRF data can provide evidence that vibrations were indeed performed by the tactors.

Limitations

There are a few limitations with the developed device. When an individual puts their foot on the ground, they still receive the normal pressure sensation from the ground. While with this device, they receive vibration stimulus on top of that pressure sense. This means the stimulation from the ground and the vibrations are different sensations. This represents an augmentation or addition to the normal sensation individuals feel during walking, opposed to a complete sensory information replacement. Finally, the lack of a check during the trial for if the tactor control box has disconnected or not is a limitation that the current system cannot control for. Implementing a check within the software that can determine if the connection with the control box has been lost could decrease the chance of this happening.

Conclusions

This study demonstrates one particular use of an online control system that performs stimulation based on an individual's movement. Many similar control systems have been developed predominately in ExoSkeleton or ExoSuit studies (Han et al. 2022, Li et al. 2023). The current control system is not limited to plantar vibro-tactile control, it can be applied for gait linked mastoid process stimulation (Chien et al. 2016), virtual reality perturbations (Eikma et al. 2016), electrical stimulation (Zehr et al. 2014), or other environmental perturbations (Roeles et al. 2018). By having a control system that stimulates, augments, or perturbs sensory systems according to an individual's movement

in real-time, would allow for a more comprehensive elucidation of the sensorimotor apparatus in health and pathology and consequently, its assessment and rehabilitation. In the next chapter, this system will be implemented in an investigation to better understand the role of the plantar surfaces in gait and balance control.

Chapter 4: Effect of Vibro-Tactile Stimulation Sequence and Support

Surface Inclination on Gait and Balance Measures

Introduction

Many studies have established that the cutaneous receptors within the plantar surfaces of the feet are important for balance control during standing and walking (Inglis et al. 2002, Felicetti et al. 2021). Specifically, during standing they have been shown to supply feedback on the changes in pressure along the skin surfaces to keep the CoM over the BoS (Inglis et al. 2002, Kavounoudias et al. 1998). This would be done through sensing the location and movements of the CoP with respect to the CoM, aiding in the perception of the body's orientation in space (Kavounoudias et al. 1998, Roll et al. 2002). However, during walking, this CoP traverses along the foot in a repeated and predictable pattern- going from heel to the toes along the lateral border of the sole (Nurse & Nigg 2001). The CoP is even adjusted during gait to keep the CoM within the BoS (Hof et al. 2007). Thus, the perception of this CoP movement may be vital to the central nervous system (CNS) for maintaining balance during walking.

One group that can aid in understanding what occurs when this sense from the plantar surfaces is removed is peripheral neuropathy patients. A prominent symptom of hyperglycemia from diabetes is a loss of tactile sensation in the soles of the feet (Alam et al. 2017). Due to this loss of sensation, people with diabetic neuropathy are at a far greater risk of falling than people with plantar sensitivity (Alam et al. 2017, Cavanagh et al. 1992). There has also been a significant relationship between plantar sensitivity of the forefoot region and scores on clinical mobility measures (Cruz-Almeida et al. 2014). This increased risk of a fall and decreased mobility may be due to the removal of the plantar

surfaces aid in the perception of the CoP movements. Interestingly, when regions of the plantar surfaces are desensitized in healthy individuals, the majority of pressures are shifted away from desensitized regions and towards regions that remain sensitive to tactile stimuli (Nurse & Nigg 2001). This could be a corrective measure in gait to maintain an accurate perception of the CoP.

If the perception of the CoP movement along the plantar surfaces was used during gait, then having the ability to give additional feedback on this movement may further aid in balance control during walking. And conversely, perturbing strictly the perception of this CoP movement would negatively affect balance during gait. One such method could be with vibrations. Vibrations stimulate specific mechanoreceptors that are sensitive to a dynamic or moving stimulus (Stzalkowski et al. 2018), possibly like the movement of the CoP. During standing, vibrations applied to regions of the plantar surfaces led to individuals leaning away from such stimulus (Kavounoudias et al. 1998), with a higher frequency increasing this effect (Kavounoudias et al. 1999). It was suggested by the authors that these high frequency vibrations induced a perception change of the CoP location. Specifically, the feeling of the CoP shifting towards the locations of the vibrations, thus leaning away was a corrective measure to shift the CoP back to the original position (Kavounoudias et al. 1998 & 1999).

Applying vibrations to the plantar surfaces during walking has been done before, usually by seeing the effects of plantar stimulation on spatiotemporal measures (Chien et al. 2017, Mukherjee et al. 2015, Novak & Novak 2006), kinematic and kinetic measures (Liang et al. 2022, Pathak et al. 2022, Song et al. 2022), and variability measures (Chien et al. 2017, Galica et al. 2009, Stephen et al. 2012, Yamashita et al. 2021). To the best of

our knowledge, there have been no studies investigating how patterned vibro-tactile stimulation on the plantar surfaces alter balance measures during gait (Xie et al. 2023).

It is important to note that walking is predominantly controlled through the sense of vision (Torres-Oviedo & Bastian 2010). Therefore, the presence of reliable visual information could minimize or completely negate the effects of applying vibro-tactile stimulation to the plantar surfaces. However, according to the multisensory integration model, when one sensory feedback mechanism becomes unreliable or is decreased, then the system increases its reliance on the remaining sensory systems (Eikema et al. 2016, Peterka & Loughlin 2004). This increased reliance on different sensory systems can also change depending on the task (Eikma et al. 2016). One task that may increase the reliance on the tactile system is inclined walking. When walking on an incline there is an increased risk of a slip due to an increase of shear forces (Sun et al. 1996). Therefore, if stimulating the plantar surfaces alters balance control, then performing the same test during inclined walking may cause a greater effect, especially when reliance on tactile sensation was increased due to low visual information.

The goal of this study will be to investigate if the sequence of stimulation that traverses across the plantar surfaces are used in a feedback model described in **Figure 9**. We hypothesize that the sequence of stimulation across the plantar surfaces, provided by the CoP movement, is a component of the predicted and actual sensory feedback. If this is the case, then we would suspect having the actual feedback not mimic the predicted feedback, by providing a stimulation sequence in an abnormal pattern, would lead to sensory error and result in balance deficits. Additionally, this will test how vibro-tactile

stimulation on the plantar surfaces alters the actual sensory feedback provided during walking.

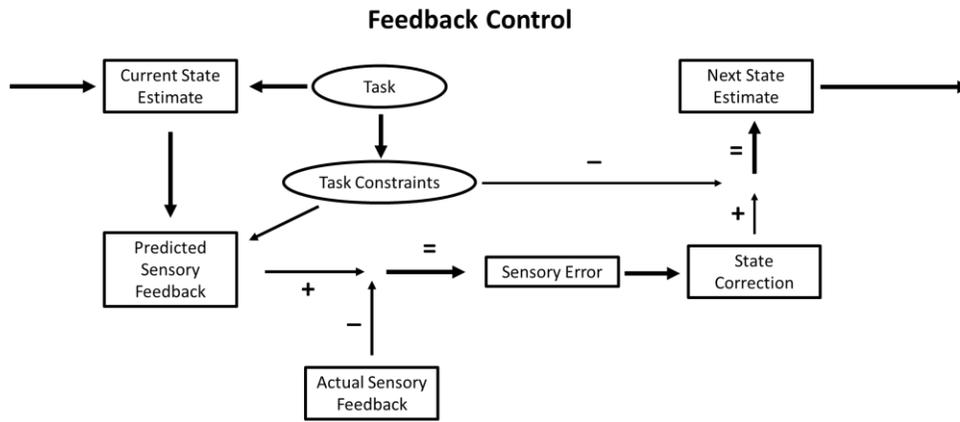


Figure 9: General feedback sensorimotor control adapted from *van Beers et al. 2002*. To reach a desired state, an internal model of the current state must be compared to the actual current state. Then using the difference of those predicted and actual body states, create corrections for the current body state to reach the desired next body state. The task at hand directly influences what this state must be. Task constraints emerge from the task and will limit the found state correction such that it fits the task. These task constraints would also affect the expected feedback- through influencing the predicted sensory feedback. For the current experiment, these task constraints are changed by the incline of the treadmill. The different stimulation patterns could influence the actual sensory feedback obtained in this cognitive level model. The immediate effects of experiencing a pattern of stimulation may lead to a decrease (Gait-Like Stimulation) or increase (Abnormal Stimulation) in the amount of sensory error the system receives. We hypothesize that an increase in sensory error would result in a larger degree of state correction, and thus result in balance deficits. Alternatively, if no balance deficits are present it could be representative of a healthy system being able to perform such state corrections effectively.

Therefore, the purpose of this study was to investigate the effects of different sequences of vibro-tactile stimulation to the plantar surfaces on spatiotemporal and balance measures during different inclines of slow walking in healthy adults. We

hypothesized that stimulation in an abnormal sequence (random stimulation) during gait, would result in gait (stance times, stance lengths) and balance deficits (stride width, foot placement, and margins of stability) when compared to stimulation that follows a natural sequence during walking. Alternatively, a lack of significant differences would indicate the healthy human ability to adjust and reweight, through multisensory integration and residual sensory feedback, such that gait and balance outcomes show minimal or no deficits. Additionally, we hypothesized that walking on an inclined surface would increase the effects of the tactile stimulation sequences on these measures when compared to walking with no stimulation.

Methods

Subjects

For this study a total of nine healthy adults (4 male, 5 female, age: 26.2 ± 3.5 , height: 166.7 ± 7.8 cm, weight: 62.4 ± 11.7 kg) were collected. These individuals were between the ages of 19 and 30 years of age, with an exclusion criterion of the presence of any disfunction including physical impairments, neurological disease, cardiovascular disease, or other abnormalities that may affect walking on a treadmill. Each participant gave informed consent prior to their participation. Ethical approval was provided by the institutional review board from the University of Nebraska Medical Center.

Equipment

All participants prior to walking performed a set of pre-tests to determine plantar sensitivity. This included Semmes-Weinstein Monofilaments (North Coast Medical Inc. Morgan Hill, CA) and 120Hz Biothesiometer (Bio-Medical Instrument Company,

Newbury, OH). These tests were done to inspect the perception sensitivity to pressure and vibrations of the plantar surfaces.

Subjects were then given a pair of Nike Free minimalist shoes with custom-made tactor embedded insoles (**Figure 11**). Each insole was fitted with six C-2 tactors (Engineering Acoustics Inc.; EAI, Casselberry, FL), placed in sets of two under the heel (heel set), base of the fifth metatarsal (MT5 set), and base of the first metatarsal and big toe (MT1 set). These tactors were set to vibrate at a constant frequency of 250Hz (Chien et al. 2017, Mukherjee et al. 2015) and maximum amplitude of 23.5db (~0.2mm). This amplitude was used so we were confident in the consistency of amplitude throughout the stimulation period, based on previous testing (**Supplementary Figure II**), while the frequency was chosen to follow previous studies (Chien et al. 2017, Mukherjee et al. 2015, Xie et al. 2023). Two tactor controlling boxes, supplied by EAI, were attached to the lower back of the subjects via a fanny pack, such that there was one box controlling the tactors within each shoe. These boxes were controlled through a custom-made MATLAB software (see previous chapter). In brief, this code allowed for real-time control of individual tactors within each shoe, to match the vibro-tactile stimulation with the gait kinematics within the stance phase of each subject.

With such control of the stimulation, subjects walked while experiencing three different stimulation patterns: No Stimulation (NS), Gait-Like stimulation (GS), and Random stimulation (RS). NS was treated as the control condition, with no tactors being activated. The GS involved activating each set of tactors within the shoes sequentially from the heel set during heel-strike to midstance, MT5 set during midstance to heel-lift, and MT1 set during heel-lift to toe-off. This was meant to follow the progression of the

normal CoP progression along the plantar surface during walking (**Figure 10**). The RS caused a random sequence of the three sets to be activated sequentially in random durations during the stance phase of gait, but not following the progression of the stance phase. This meant that tactor sets activated in a perturbing pattern, against the normal CoP progression and not according to the real-time movements.

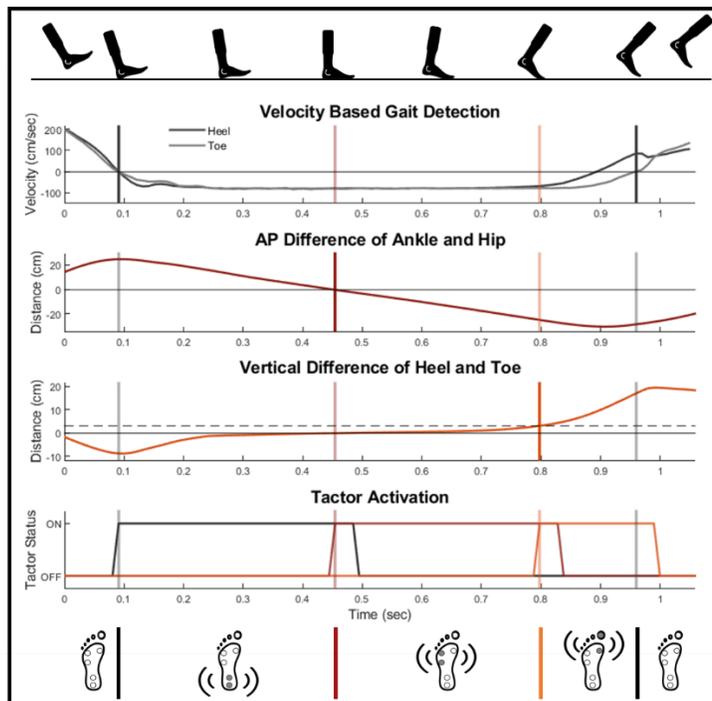


Figure 10: GS control system takes kinematic measures during gait to activate specific tactor sets that follows the natural progression of walking. Stance is determined by the velocity of the heel and toe markers (Zeni et al. 2008). Then midstance and heel lift is found by finding the AP position of the ankle and hip, and the vertical height of the heel and toe markers, respectively. The last two rows show the tactor set activations based on the gait event detection points in the first 3 rows. Colored bars indicate the specific detection events that activate the specific tactor sets.

Participants walked on a force plate instrumented split belt treadmill (Bertec Version2.0 2013, Columbus, OH) that collected GRF at 1000Hz. Marker position data

was collected using a 16-camera motion capture system (Vicon Oxford, UK) at 100Hz. Reflective markers were placed on bony landmarks following the PlugInGait Full-Body AI from Vicon (**Figure 11**).



Figure 11: Experiment and equipment set-up for data collection. (**Left**) Custom made tactor embedded insoles that fit into the specific shoe size of each subject. These tactors were then connected to the tactor boxes that were attached to a fanny pack by Velcro. Subjects wore sunglasses in the dark room to decrease visual information and increase reliance on tactile feedback. (**Right**) Subjects wore the fanny pack around their waist such that the tactor boxes were on their back above the posterior pelvic markers.

Procedure

Before subjects began walking, they went through two familiarization trials. The first was a familiarization trial to remove any surprise effects from when the tactors first activate during the walking trials. This was done by having the subjects stand as each tactor set between the two feet were activated for 0.3secs one at a time in random order. The subjects were asked to state which tactor set was activated using a key showing the tactor set locations (**Supplementary Figure III**). An additional benefit of this familiarization trial was to enable the subjects to distinguish the locations of the different tactor sets, and thus to distinguish the feeling of the different stimulation patterns. For the

next familiarization trial subjects performed a 5min walk to habituate to the conditions of low light and sunglasses during level walking at 0.8m/s.

Participants performed two trials of each incline at 0.8m/s (Moore et al. 2015), with the order of incline being randomized. This resulted in six total trials and 35mins of walking, with at least 2mins break between trials. Each trial consisted of 5mins of walking, within each trial, subjects experienced 1min of each stimulation pattern in a randomized order. These stimulation sections of the trial were separated by 30secs of NS (Baselines) before and after each pattern, which was treated as a time for the subject to return to a baseline walking pattern before the next stimulation pattern began (**Figure 12**). This resulted in two minutes of walking data (about 90 steps on average) for each participant in all condition combination.

After subjects completed all trials, we wanted to ensure that any gait changes we may see could not be attributed to pain or discomfort from the vibrations or presence of the tactor embedded insoles (Rossignol et al. 2006). For this we had subjects provide a visual analog scale (0-10) rating of the general comfort of the insoles, with 0 being the least and 10 being the most comfortable. Then we asked if that rating changed when the tactors were active. Finally, we wanted to know if subjects were able to notice the different stimulation patterns while walking. Subjects were not told before participating about the different stimulation patterns; thus, this was a test of how attentive individuals were in sensing a moving vibratory stimulus on the plantar surfaces during walking. This was done by asking if they felt anything different between the two moments of vibration during a walking trial at the end of the final trial.

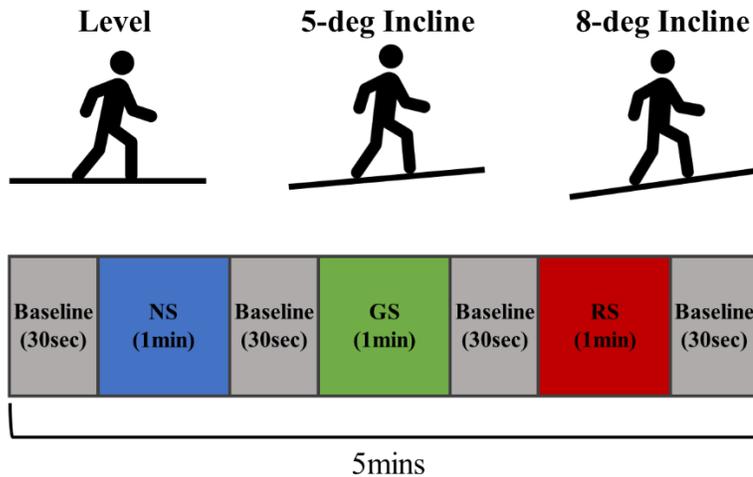


Figure 12: Protocol of one trial that was randomly repeated at each incline. Individuals walked for a total of 5mins where 1min of each stimulation pattern was experienced. There were 30sec breaks of no stimulation between the three patterns to allow the subject to return to a normal baseline of walking before the next stimulation.

Data Analysis

Data analysis was performed in MATLAB. Due to individuals walking with low visual information, step overs onto the contralateral belt were common. For this reason, gait events were found using the velocity of heel and toe markers of each foot. A heel strike was defined as a heel AP velocity change from positive (forward) to negative (backward), and the opposite change for the toe velocity (Zeni et al. 2008).

Spatiotemporal measures such as stance time, stance length and stride width were calculated from these gait events. Stance time was the duration of a heel strike to the following toe off of the ipsilateral side, and stance length was the limb excursion of the foot across the belt (Hoogkamer et al. 2013) normalized to the body height of each

subject. Stride width was the mediolateral (ML) distance between the heel markers of both feet at each heel strike.

Foot placement was the mediolateral and anteroposterior position of the heel marker at heel strike with respect to the position of the CoM. Foot placements were then analyzed further with a 95% confidence ellipse. This ellipse was made by finding the eigenvectors that describe the locations of each foot placement for the duration of a stimulation pattern (1min). These eigenvectors make up the direction of the major and minor axes of the ellipse. The radii of the ellipse were found by multiplying the square root of the eigenvalues by the Chi square value of 2.4477 that represents a 95% confidence interval. Then these radii were multiplied by a rotation matrix based on the angles of the eigenvectors to the x-axis. Thus, resulting in a 95% confidence ellipse that is oriented according to the spread of heel strikes. The areas of these ellipses were compared between conditions (**Supplementary Figure IV**). A larger area represents more sporadic and widespread foot placements, and a small area represents more consistent foot placements.

The margins of stability (MoS) were analyzed using marker position and velocity data. MoS was calculated as the minimum distance between the base of support (BoS) and the extrapolated center of mass (XCoM) (**Supplementary Figure V**). The BoS was estimated as the position of the ankle marker (Hak et al. 2013), while the XCoM was calculated as described in **Equations 2-4**. In brief, it is the position of the CoM plus the velocity of the CoM (v_{com}), including the walking speed for the AP direction, divided by the pendulum eigen frequency (ω_o) (Hof et al. 2007). The eigen frequency is calculated as the square root of the force of gravity (g) divided by the effective height of the CoM (h),

which was 1.34 times leg length (l) (Hof et al. 2007). Position of the CoM was estimated by the average position of all the pelvic markers (ASISs, PSISs, and Sacrum).

$$\text{Equation 2:} \quad XCOM = x_{CoM} + \frac{v_{CoM}}{\omega_{CoM}}$$

$$\text{Equation 3:} \quad \omega_{CoM} = \sqrt{g/h}$$

$$\text{Equation 4:} \quad h = (1.34)l$$

The MoS was found in the ML and AP directions at different times points of the gait cycle. The ML MoS was calculated as the minimum distance between the XCoM and BoS throughout the stance phase of gait (Hoff et al. 2007, Hak et al. 2013). The AP MoS was found at the moment of heel strike and midstance (Vieira et al. 2017ab, Young et al. 2012, Peebles et al. 2016). Midstance was defined as the moment of the ankle marker becoming in line with the CoM.

Statistical analysis was performed in SPSS 16.0 (IBM Corporation, Armond, NY). To test significant differences between the effects of inclines and stimulation patterns, a 2-way 3x3 repeated measures ANOVA (Level/5Incline/8Incline x NS/GS/RS) was performed with a significance level of 0.05. If significant differences were found a Tukey post-hoc was done for finding directionality of differences.

Results

Sensory Perception

The sensory thresholds of the subjects were within normal ranges. With the monofilament test averaging around a size 4 filament (<1.4g of force), and vibration perception within 0.05microns amplitude at 120Hz (Mosby et al. 1995, Gregg 1951).

Monofilament Test												
Subject	Right (Filament)						Left (Filament)					
	Big Toe	MT1	MT5	Sole	Heel	Little Toe	Big Toe	MT1	MT5	Sole	Heel	Little toe
1	4.31	2.83	4.31	3.61	4.31		4.31	2.83	4.31	3.61	4.31	
3	3.61	4.31	3.61	3.61	4.31		3.61	3.61	4.31	4.31	4.31	
4	4.31	2.83	4.31	4.31	4.31	4.31	2.83	4.31	2.83	2.83	4.31	4.31
5	3.61	3.61	3.61	3.61	4.31		3.61	3.61	3.61	3.61	4.31	
6	2.83	4.31	3.61	4.31	3.61	3.61	3.61	3.61	4.31	3.61	4.31	4.31
7	4.31	4.31	4.31	4.31	4.31	4.31	4.31	4.31	4.31	4.31	4.31	4.31
8	3.61	2.83	4.31	2.83	3.61	3.61	3.61	2.83	4.31	2.83	3.61	3.61
9	4.31	3.61	4.31	3.61	4.31	4.31	4.31	3.61	3.61	3.61	4.31	4.31
11	3.61	3.61	4.31	4.31	3.61	4.31	2.83	3.61	4.31	4.31	4.31	3.61
average:	3.834444	3.583333	4.076667	3.834444	4.076667	4.076667	3.67	3.592222	3.99	3.67	4.232222	4.076667
std. dev:	0.514177	0.641171	0.35	0.514177	0.35	0.361478	0.576021	0.523684	0.530189	0.576021	0.233333	0.361478
mode:	4.31	2.83	4.31	3.61	4.31	4.31	3.61	3.61	4.31	3.61	4.31	4.31

Biothesiometer Test						
Subject	Right (microns)			Left (microns)		
	MT1	MT5	Heel	MT1	MT5	Heel
1	0.01	0.01	0.04	0.01	0.04	0.04
3	0.16	0.04	0.09	0.09	0.04	0.16
4	0.04	0.04	0.04	0.04	0.04	0.04
5	0.04	0.04	0.04	0.09	0.04	0.04
6	0.04	0.04	0.04	0.04	0.04	0.04
7	0.16	0.09	0.09	0.09	0.04	0.09
8	0.04	0.04	0.04	0.04	0.04	0.04
9	0.09	0.04	0.04	0.09	0.04	0.09
11	0.04	0.04	0.04	0.04	0.04	0.04
average:	0.068889	0.042222	0.051111	0.058889	0.04	0.064444
std. dev:	0.055553	0.02048	0.022048	0.031002	0	0.041866

Figure 13: Sensory perception of pressure supplied by monofilaments (**Top**) and vibration (**Bottom**). The ranges of filaments throughout the different foot regions with the MT1, Big toe, and foot sole as the most sensitive. Blank boxes represent missing data. The MT5 region seemed to be the most sensitive to vibrations.

Spatiotemporal

There were no significant main or interaction effects from incline and stimulation pattern on stance time (Incline: $F=1.148$, $p=0.322$; Stim Pattern: $F=0.159$, $p=0.854$), stance length (Incline: $F=0.676$, $p=0.440$; Stim Pattern: $F=0.14$, $p=0.87$), or stride width (Incline: $F=0.54$, $p=0.593$; Stim Pattern: $F=0.678$, $p=0.522$).

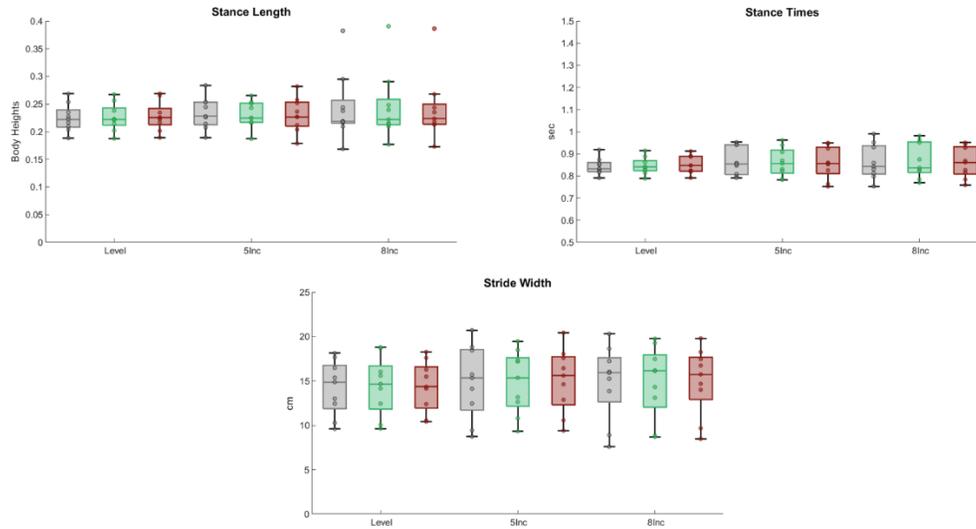


Figure 14: There was no significant effect on spatiotemporal measures from stimulation pattern or incline.

Balance Measures

There was no main effect of the stimulation pattern on any of the balance measures, MoS's and foot placement area. However, there was a main effect of incline on Foot placement area ($F=7.849$, $p=0.004$), AP MoS measures at heel strike ($F=163.925$, $p<0.001$), and AP MoS at midstance ($F=58.264$, $p<0.001$). Foot placement significantly increased by about 14cm^2 when going from level walking to inclined walking. However, there was no significant difference in foot placement area when transitioning from 5deg to 8deg of incline. For AP MoS at heel strike, as incline increased there was a significant increase of about 10.8cm from level to 5deg, and an increase of about 6.2cm from 5deg to 8deg. Lastly, for AP MoS at midstance, there was a significant decrease of about 2.2cm from level to 5deg, and a decrease of 4.3cm from 5deg to 8deg.

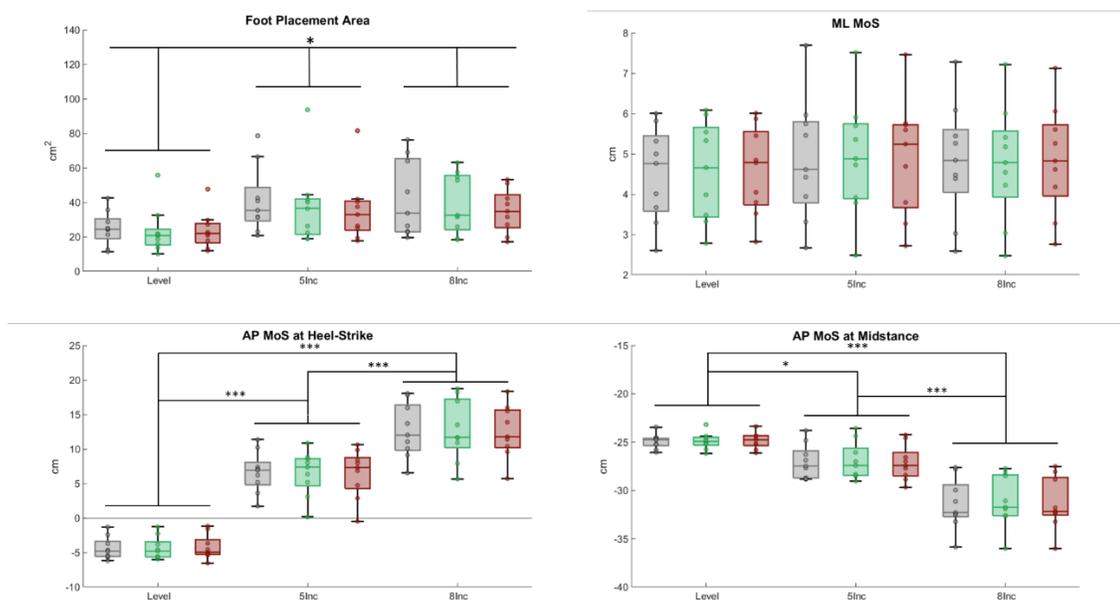


Figure 15: Stimulation patterns had no significant effect on balance measures. However, walking incline resulted in significant changes in Foot placement area, AP MoS at heel strike, and midstance. (* indicates $p < 0.05$; *** indicates $p < 0.001$)

Comfort and Pattern Perception

Comfort Scale				Pattern Responses	
Subject	OFF	ON	Difference	Response Examples	Frequency of Response
1	6	5	-1	"The stimulation felt stronger sometimes"	2
3	5	6	1	"There was a forward sequence and a backward sequence"	1
4	7	6	-1	"There were different patterns"	1
5	4	7	3	"No difference"	5
6	4	4	0		
7	9	4.5	-4.5		
8	5	4.5	-0.5		
9	7	8	1		
11	6	6	0		
average	5.888889	5.666667	-0.2222222		
std. dev	1.615893	1.299038	2.0327185		

Figure 16: Post-test results of comfort scale of insoles (**Left**) and if subjects perceived the different patterns of stimulation (**Right**). Comfort was average on the 10-point scale, with an overall small effect of active vibrations. Most individuals were unable to perceive any difference from the stimulation patterns. Only one person noticed them, while others thought the strength of the vibrations was altered.

Discussion

In the current study we investigated the effects of different sequences of vibrotactile stimulation on the plantar surfaces during the stance phases of gait. This was done at different walking inclines, and with low visual information to increase the reliance on the tactile system. We found a lack of significant effects from stimulation patterns in both spatiotemporal and balance measures; only incline led to a change in some balance measures. This gives support to our alternate hypothesis that healthy humans have the ability to adjust and reweight, through multisensory integration and residual sensory feedback, such that gait and balance outcomes show minimal or no deficits.

The effect of incline on balance measures

Incline had an effect on the AP balance measures, but not the ML balance measures of MoS. Firstly, the decrease in AP MoS at midstance as incline increases has been shown previously (Vieira et al. 2017b). However, this decrease in previous studies was thought to be caused from a decrease in stride length, while there were no significant changes in stance length for the current study (**Figure 14**). We suggest the decreases in AP MoS at midstance demonstrates an anticipation effect for the increase of AP MoS at heel strike during inclined conditions. As incline increases the body must exert more energy to overcome the increased force of gravity to progress forward (Sun et al. 2010). This increase of AP MoS at heel strike demonstrates that at the moment of heel strike the acceleration of the CoM to progress to the next step is performed later in the stance phase, most likely during push-off. Whereas during level walking, the AP MoS at this time is negative, suggesting that the CoM has already accelerated to advance to the next step. This could demonstrate the mostly passive nature of walking on a level surface. So,

it could be the AP MoS is decreasing by leaning into the next step a little sooner to slightly decrease the effort performed during the push-off phase.

Concerning the ML MoS, previous studies have shown an increase in ML MoS during inclined walking (Vieira et al. 2017ab). However, these studies had individuals walk at their preferred walking speed, as opposed to a set slow walking speed in this study. Walking at a slow pace increases ML MoS (Suptitz et al. 2012) and putting an individual in unstable walking scenarios also increases MoS (MacDonald et al. 2022). Thus, no effect of incline may have been seen in the current study because individuals were already walking at an increased MoS from the slow walking speed and the low visual information from the dark room and sunglasses. This could also explain the lack of an effect incline had on spatiotemporal measures.

Vibro-tactile stimulation had no effect on gait measures

Different stimulation patterns led to no significant changes to any of the variables tested in this study. These results contradict a previous study investigating the effects of similar stimulation during different walking inclines (Xie et al. 2023). Where the current study differs is the timing and patterns of stimulation. Most vibro-tactile stimulation studies have the vibrations present throughout the entire gait cycle - including the swing phase (Xie et al. 2023, Chien et al. 2017, Mukherjee et al. 2015). The timing of stimulation used in the present study occurred only during the stance phase of gait, when the plantar surfaces would actually be supplying information about the environment. This is because unlike the hand, the mechanoreceptors within the plantar surfaces do not have any passive activations with the absence of pressures, such as the swing phase of gait (Kennedy et al. 2002). Thus, if the plantar surfaces are being stimulated with vibrations

during swing it would not be an augmentation of sensory feedback, but a completely new perturbing sensation. We suggest the current study demonstrates that supplying vibrations to the plantar surfaces during the stance phase may lead to differing effects than stimulating throughout the entire gait cycle.

Vibro-tactile stimulation may not affect the perception of the CoP

In the current study we attempted to alter the perception of the CoP movement similar to a postural study that applied high frequency plantar vibrations (Kavounoudias et al. 1998, 1999). These studies found whole-body shifts away from vibro-tactile stimulation applied to the plantar surfaces, due to a perceived shift in the CoP due to the mechanoreceptor stimulation. However, in the present study, similar vibrations had no effect on gait measures. There are two main possibilities: (1) the plantar surfaces are not involved in the perception of CoP movement during gait, or (2) vibrations at 250Hz do not alter the perception of the CoP.

It is possible that the time delay from tactile stimulation to its perception in the brain or spinal cord is too long for reliable balance control. By the time the brain learns how or where the CoP is moving, the system may already be within the next step. However, it has been shown that the CoP is shifted medially or laterally during a stance phase to maintain a stable ML MoS (Hof et al. 2007). Thus, it would make sense for the plantar surfaces to supply the afferent signals to complete a feedback loop of motor control (van Beers et al. 2002), similar to simple reflexive feedback control such as withdraw reflexes (Sherrington 1910). Additionally, individuals walked at a slow walking speed, resulting in about 0.8sec stance time, which is sufficiently long for afferent signals of the foot to reach the CNS for processing.

Additionally, there have been studies that show a strong neural connection between the motor control centers of the brain and the sensory representations of the foot. Activations of motor control centers, such as the supplementary motor cortex, can occur just by stimulating the plantar surfaces in a gait-like sequence (Zhang et al. 2019, Labriffe et al. 2017). These studies suggest that these coactivations of sensory and motor areas to be evidence of plantar tactile feedback being used in gait control. Therefore, while there was no effect of plantar stimulation found in the current study, it may not necessarily mean that the plantar surfaces are not being used for balance and gait control. It is possible that in the sufficiently long stance phase, multisensory integration allows adjustments of sensory weights such that balance and gait outcomes show minimal effects.

The second possibility is that the vibrations we supplied to the plantar surfaces do not alter the perception of the CoP, and thus did not alter the gait measures tested. This could indicate that healthy individuals are able to distinguish the sense of pressures applied to the foot from the ground and the vibrations supplied by the tactors. Fast adapting (FA) fibers are most sensitive to vibrations and moving stimuli across the surface of the skin (Stzalkowski et al. 2018). This led us to believe that these FA fibers may aid in perceiving the moving CoP during stance. Thus, the addition of vibrations would negatively impact the signal to noise ratio and result in gait related effects. However, SA fibers are known to feel pressure and give information on the level of pressures applied to the skin while not being sensitive to vibrations (Zimmerman et al. 2014, Gardner 2010). The rate of action potentials sent by these SA fibers reflect the amount of pressure applied to the skin (Zimmerman et al. 2014). Therefore, different

regions of the foot would supply a higher rate of action potentials at different moments of the stance phase. Healthy individuals may be able to make do with this reduced signal to noise ratio from the FA fibers during stimulation because it may not be the main sensory fiber type the brain is using to sense the CoP movement.

Additionally, the addition of vibrations altering the signal to noise ratio for sensing the CoP, may only lead to changes in behavior if the task allows it. During standing individuals can adjust the CoP throughout the BoS. However, during walking, the CoP movement is a result of performing limb progression during stance - to move on to the next step. Walking may require a much larger decrease in this signal to noise ratio than standing to lead to an emerging effect in behavior due to this requirement. The healthy individuals may have received conflicting sensory information from what they predicted, resulting in a larger correction to their gait control, however, they were successfully able to make this correction- maintaining proper balance (**Figure 17**). Thus, to properly test if the CoP movement is used for balance control during walking, a more selective and detailed method of stimulation or task must be used.

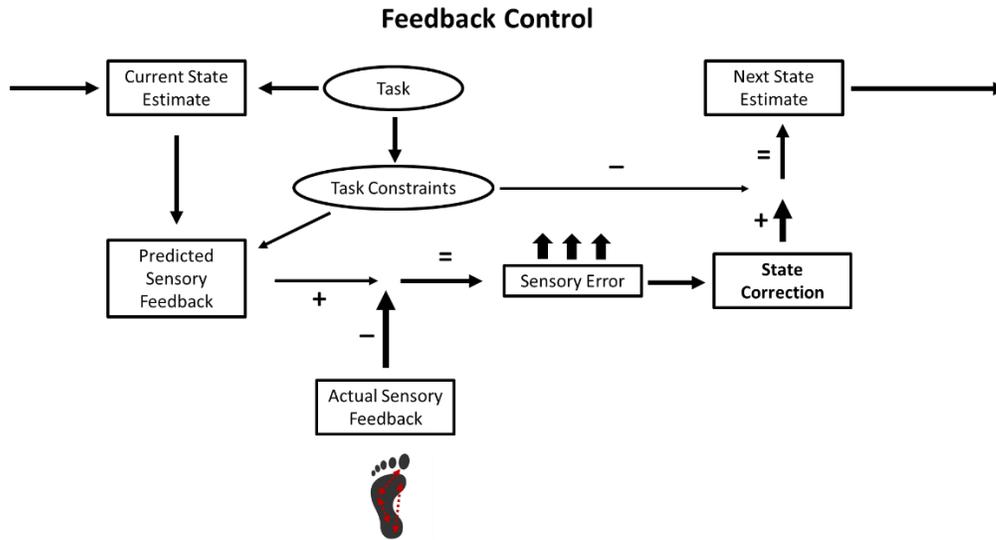


Figure 17: Feedback model revisited with the possible effect of RS. Giving abnormal stimulation during gait must have altered the actual sensory feedback they received during the task, leading to an increase of perceived sensory error. This may have led to the system creating a state correction to remain in balance, resulting in the lack of effect in emergent behavior through the collected balance measures. This stimulation may demonstrate the flexibility and adaptability of a healthy system.

Limitations

This study comes with some limitations, firstly some trials had to be dropped out of the analysis because of a disconnection with the tactor control box. When validating the activations of tactors during trials, it was found that the tactor controller boxes and the computer would be disconnected for either the whole or parts of the 5min trial. For this reason, only nine participants were used in the analysis of the current data set, out of originally collecting 14 participants. Additionally, of these nine participants, only four had two full successful trials at each incline. The remaining had either two or one trial at a particular incline used within the data analysis. Unfortunately, there was no way during data collections to know if connection with the tactor box was lost or not, and it could

only be found after the trial. This is another reason why we suggest for future studies to validate the vibrations supplied by the tactors.

It could also be that inclined walking was not the best way to increase the reliance on tactile feedback. A previous study found that declined and inclined walking led to a stronger effect from supra-threshold tactile stimulation (Xie et al. 2023). However, this stimulation was provided throughout the entire gait cycle, including the swing phase. Thus, the effects found from that study may have been from the perturbing sensation of vibration during the swing, opposed to an augmentation of sensation while the foot was on the ground (see previous Chapter's Introduction). Future studies should investigate if specifically stimulating the plantar surfaces during swing produce previously found effects (Xie et al. 2023).

Next, the estimation of the CoM was performed through pelvis markers opposed to the full body marker set. Previous studies have used pelvis markers as a good estimation of the CoM for similar MoS measures as shown in the current study (Buurke et al. 2023, Hak et al. 2013). The inclusion of the upper body in the CoM position may have added more accuracy to the exact CoM position and velocity. However, due to participants walking normally at a slow speed, we do not expect this increased CoM estimation to lead to large changes in the seen MoS values.

Finally, the tactors used in this study were susceptible to vibration dampening when pressure was applied to them. We added a spacer to the tactor to decrease this dampening effect (see previous chapter) however, this only decreased the effect it did not remove it. Therefore, the amplitude and sensation of the vibrations may not have been consistent. In fact, some subjects thought the strength of the stimulation was being

altered, as opposed to the sequencing of activations (**Figure 8**). During the GS, factors were activated to follow sequentially the regions with the most pressure on the foot; thus, this feeling of stronger vibrations was most likely during the RS when the vibrations did not follow this pressure. While this effect was unwanted, we do not believe it diminished the stimulation effects.

Conclusion

In the current study we investigated how different sequences of vibro-tactile stimulation altered spatiotemporal and balance measures during level and inclined walking at low vision. However, very little effects of stimulation patterns were found. Therefore, healthy humans have the ability to adjust and reweight, through multisensory integration and residual sensory feedback, such that gait and balance outcomes show minimal or no deficits when foot-sole tactile sensory sequences are manipulated in low vision conditions especially during slow walking conditions. It is possible that the perception of CoP movement is supplied through SA mechanoreceptor fibers that are not typically sensitive to vibrations. This work gives indication to the flexibility and adaptability of a healthy motor control system and demonstrates a method of testing such a system with an online stimulation control software. It remains to be seen in individuals with sensory deficits of the foot, whether the specific sequence of augmented tactile stimulation could improve gait and balance metrics.

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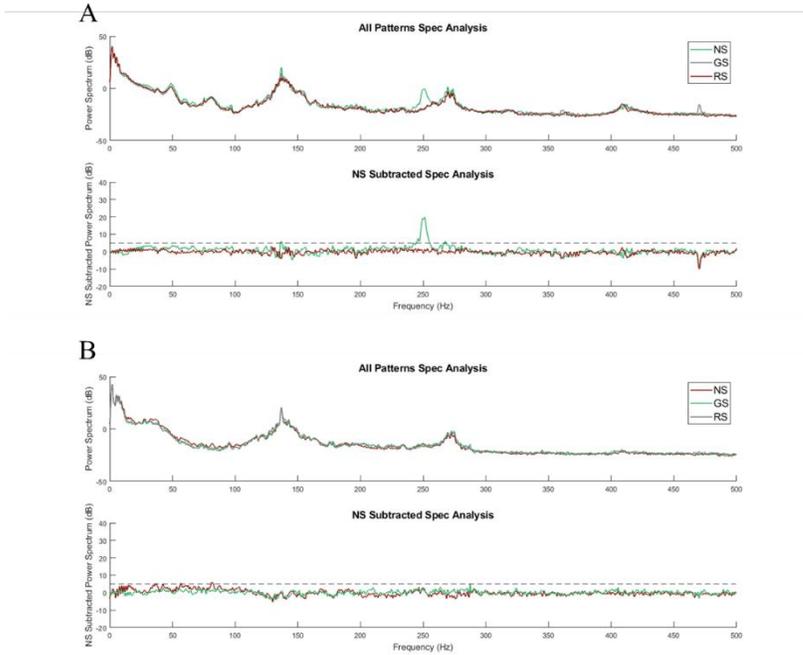
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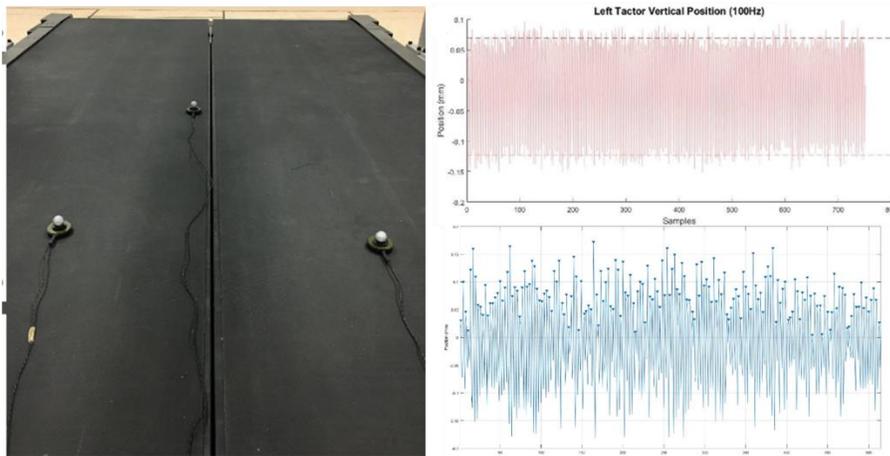
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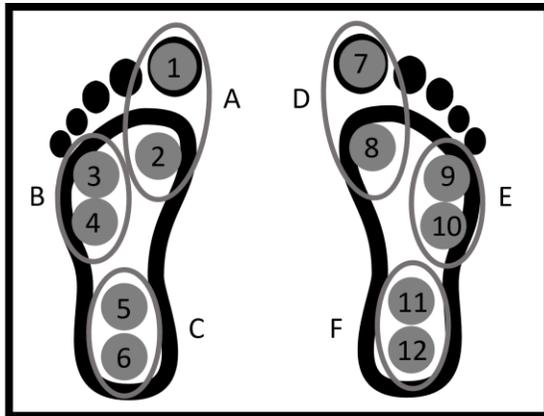
Appendix



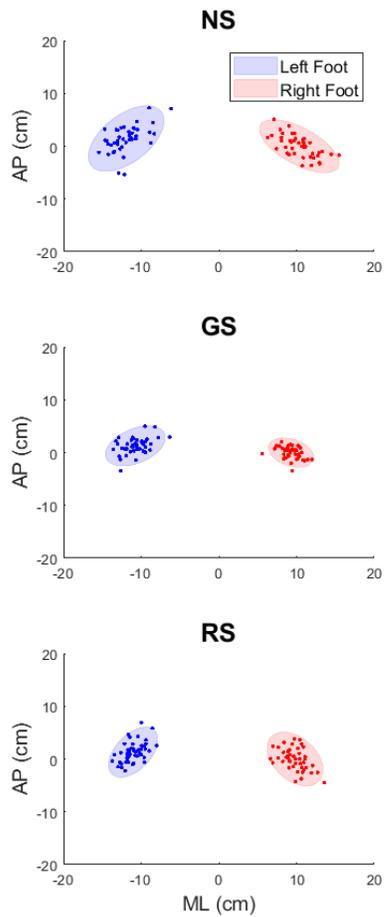
Supplementary Figure I: Examples of failed stimulation trials, see **Figure 6** for further detail of graphs. **(A)** Spectral analysis of trial where tactor control boxes disconnected during a trial. Tactors were active at 250Hz for GS but disconnect before RS began. **(B)** Trial where tactor activation signals were sent to the tactor box, but boxes became disconnected; leading to no spectral peak at 250Hz and thus a lack of vibrations supplied to the plantar surfaces.



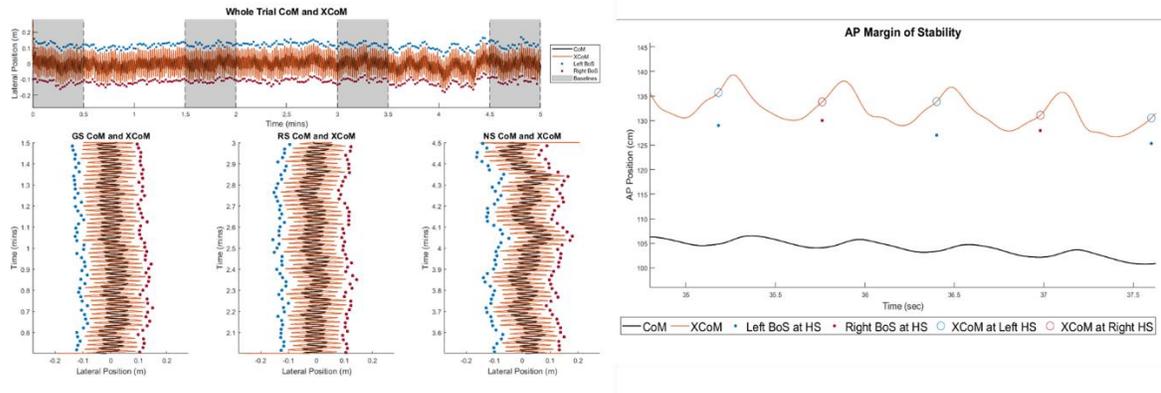
Supplementary Figure II: A marker was placed on the mechanical vibrator of the tactors (**Left**) to record marker movement at 300Hz during different amplitudes of vibrations. At maximum amplitude (23.5db) there is a smooth 0.2mm peak to peak amplitude. However, during a smaller amplitude (17.5db) the amplitude of the marker movements is sporadic. Thus, we used 23.5db to be confident in a consistent amplitude and strength of stimulation.



Supplementary Figure III: Subjects were given this key to answer what tactor set was being activated when during the tactor familiarization. Subjects were asked to respond with the what lettered circle was activated.



Supplementary Figure IV: Representative example of foot placement area calculation. Each colored dot represents the location of the heel, with respect to the CoM, at the moment of heel strike. Each data point was shifted down by the overall average position of both limb's foot placements. This was done so the y-axis would be the same for each participant, and such that the orientation of both limb's foot placements was not altered. Then a 95% confidence interval ellipse is drawn to represent the spread of these foot placements for both feet. The area of the left and right ellipses was averaged and then compared between conditions.



Supplementary Figure V: Examples of XCoM and MoS calculations. **(Left)** The XCoM, CoM, and both sides BoS are shown throughout an entire trial, as well as broken down to the individual stimulation pattern sections. The ML MoS was determined by the distance between the peak of XCoM and the BoS for that stance period. **(Right)** Depiction of AP XCoM, CoM, and both sides BoS. The XCoM is constantly ahead of the CoM because the individual is walking at a speed of 0.8m/s in that direction. The AP MoS at heel strike was calculated as the distance between the BoS at heel strike (solid dot) and the XCoM at the moment of heel strike (open circle) of the same color. The same calculation was done for AP MoS at midstance but that distance at the moment of midstance instead.

		NS	GS	RS
Stance Time (sec)	Level	0.843 (0.037)	0.845 (0.038)	0.849 (0.040)
	5Inc	0.866 (0.065)	0.862 (0.065)	0.859 (0.073)
	8Inc	0.863 (0.078)	0.868 (0.080)	0.860 (0.070)
<hr/>				
Stance Length (body heights)	Level	0.225 (0.025)	0.226 (0.025)	0.228 (0.026)
	5Inc	0.233 (0.029)	0.232 (0.025)	0.231 (0.031)
	8Inc	0.243 (0.062)	0.245 (0.063)	0.241 (0.060)
<hr/>				
Stride Width (cm)	Level	14.20 (3.076)	14.45 (3.312)	14.39 (2.845)
	5Inc	14.86 (4.139)	14.86 (3.558)	15.06 (3.601)
	8Inc	14.86 (4.209)	14.87 (4.051)	14.98 (3.800)
<hr/>				
Foot Placement Area (cm ²)	Level	25.09 (10.49)	23.38 (13.84)	22.65 (11.00)
	5Inc	41.17 (20.21)#	38.03 (23.48)#	35.86 (19.91)#
	8Inc	42.32 (23.65)#	39.51 (18.71)#	35.10 (12.80)#
<hr/>				
ML MoS (cm)	Level	4.497 (1.430)	4.574 (1.483)	4.572 (1.341)
	5Inc	4.871 (1.684)	4.920 (1.612)	4.917 (1.667)
	8Inc	4.822 (1.630)	4.763 (1.609)	4.858 (1.536)
<hr/>				
AP MoS HeelStrike (cm)	Level	-4.426 (1.747)Ψ	-4.401 (1.701)Ψ	-4.249 (1.876)Ψ
	5Inc	6.641 (3.193)#Ψ	6.486 (3.363)#Ψ	6.462 (3.681)#Ψ
	8Inc	12.71 (4.070)#	12.81 (4.583)#	12.512 (3.922)#
<hr/>				
AP MoS Midstance (cm)	Level	-24.92 (0.797)Ψ	-24.89 (0.879)Ψ	-24.86 (0.868)Ψ
	5Inc	-27.03 (1.811)#Ψ	-26.97 (1.961)#Ψ	-27.16 (1.872)#Ψ
	8Inc	-31.43 (2.761)#	-31.15 (2.800)#	-31.36 (2.814)#

Supplementary Figure VI: Results for all variables at each incline of walking and each stimulation pattern. # indicates a significant difference from level incline ($p < 0.05$). Ψ indicates a significant difference from the 8deg of incline ($p < 0.05$).