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Mastoid Vibration Affects Dynamic Postural Control During Gait

Jung Hung Chien, Mukul Mukherjee & Nicholas Stergiou

Keywords: Biomechanics, Posture, Sway variability, Nonlinear, Sample entropy

Abstract

Our objective was to investigate how manipulating sensory input through mastoid vibration (MV) could affect dynamic postural control during walking, with and without simultaneous manipulation of the visual and the somatosensory systems. We used three levels of MV (none, unilateral, and bilateral) via vibrating elements placed on the mastoid processes. We combined this with the six conditions of the Locomotor Sensory Organization Test (LSOT) paradigm to challenge the visual and somatosensory systems. We hypothesized that MV would affect both amount and temporal structure measures of sway variability during walking and that, in combination with manipulations of the visual and the somatosensory inputs, MV would augment the effects previously observed. The results confirmed that MV produced a significant increase in the amount of sway variability in both anterior-posterior and medial-lateral directions. Significant changes in the temporal structure of sway variability were only observed in the anteriorposterior direction. Bilateral MV produced larger effects than unilateral stimulation. We concluded that sensory input while walking could be affected using MV. Combining MV with manipulations of visual and somatosensory input could allow us to better understand the contributions of the sensory systems during locomotion.

Introduction

Recent epidemiological evidence estimates that approximately 30% of adults above the age of 40 might experience some form of vestibular dysfunction. <u>1</u> Such dysfunction could eventually lead to chronic dizziness and imbalance that can have significant impact on fundamental activities of daily life such as walking. <u>37</u> Therefore, it is important to be able to diagnose vestibular problems and treat them appropriately to avoid decreases in mobility and falls. However, our knowledge of how the vestibular system affects balance and dynamic postural control during walking is limited.

Few studies have investigated how patients with vestibular disorders maintain balance during walking. <u>10,14</u> The limited research has revealed that patients with a bilateral vestibular disorder were able to walk successfully blindfolded over a short distance without any lateral deviation, even though these patients were much slower than controls. However, blindfolded patients with a unilateral vestibular disorder walked with significant lateral deviations in the direction of the lesion. These findings suggest that

the contribution of the vestibular system to dynamic postural control during walking could be of great importance.

To avoid unnecessary exposure of patients with vestibular disorders to untested methodological procedures, several investigators have instead tested healthy individuals that were subjected to caloric and galvanic stimulation to study the role of the vestibular system in dynamic postural control during gait. The caloric method tests the function of the vestibular system using air or water irrigation on the external ear canals.21 With caloric vestibular stimulation, subjects showed increased lateral deviation at the hip but not at the foot, neck or head during treadmill walking with eyes open.34 In galvanic vestibular stimulation a small amount of galvanic current is applied to the mastoid process to modulate the continuous firing level of the peripheral vestibular afferents.25 This causes participants to lean in different directions during walking depending upon the polarity of the current. The effect of galvanic vestibular stimulation depends on the walking speed, 29 however, such that when walking speed increases, the effect is attenuated and the lateral deviation diminishes. Importantly, both galvanic vestibular stimulation and caloric irrigation induce discomfort.18,40 Therefore, the usability of these approaches to study the true effects of the vestibular system on balance and dynamic postural control during walking is guestionable as anxiety due to discomfort may compromise subject responses.

To overcome the limitations with these techniques, scientists have employed vibration stimulation as an alternative method of vestibular manipulation. Specifically, Karlberg *et al.* <u>32</u> showed that abnormal eye movements (nystagmus) induced by mastoid vibration (MV) are similar to those observed in patients with acute unilateral vestibular deficit. Further, vibrating the neck muscles can cause significantly more nystagmus in patients with unilateral vestibular lesion.<u>32</u> This suggests that the vestibular system is sensitive and responsive to MV.<u>43</u> Moreover, MV is considerably more comfortable than the caloric test.<u>43</u> For these reasons, MV presents a viable alternative method for the investigation of the effects of the vestibular system on postural control.

Research has shown that MV induces dizziness or unsteadiness and further influences postural control by affecting the body-centered coordination system. <u>2,7,8</u> Furthermore, in a previous study during which healthy subjects underwent *positron emission tomography* (PET) assessments whilst receiving MV, it was found that the areas of the perisylvian cortex, temporoparietal junction and somatosensory area II were the common activation regions. These areas are those involved for vestibular and neck muscle representations of body orientation in space. <u>3</u> MV was also found to affect body orientation in healthy controls but not in patients with cervical dystonia during stepping-in-place. <u>4</u> This study confirmed that neck sensation is crucial for combining the information from the vestibular system and the neck muscle spindles for controlling posture. Vestibular input cannot identify if the head or the entire body is progressing especially when the head is moving over a stationary torso. Thus, input from the neck muscles is fundamental in informing the nervous system regarding movements of the head with respect to the torso and the head yaw rotation. <u>11</u>

Current clinical testing using the Sensory Organization Test (SOT) manipulates only the visual and somatosensory inputs to study their effects on postural control during standing; the vestibular system is not manipulated. Recently, our group has developed the Locomotor Sensory Organization Test (LSOT)12,13 as a parallel to the SOT. The LSOT uses sequential manipulations of different sensory systems to study the effects of sensory inputs on dynamic postural control during walking. Our previous research with the LSOT has shown that dynamic balance control during walking in healthy individuals is affected by the manipulation of multisensory inputs. 12,13 The amount of sway variability observed during walking reflects similar balance performance with standing posture, indicating that similar feedback processes may be involved.12 However, the contribution of visual input is significantly increased during walking in comparison to standing. Our results with respect to the temporal structure of sway variability also revealed that as sensory conflict increases, more rigid and regular sway patterns are found during standing.13 The opposite is the case with walking where more exploratory and adaptive movement patterns are present. In our previous studies, 12, 13 however, vestibular inputs were not systematically manipulated. Thus, the effects of vestibular sensory inputs on dynamic postural control during walking remain unknown.

Therefore, the purpose of the present study was to combine MV with the LSOT paradigm to determine the contributions of the vestibular system to dynamic postural control during walking. Sway variability measures were used as previously described to investigate dynamic postural control. <u>12,13</u> We hypothesized that the MV would affect both the amount and the temporal structure of sway variability during walking and, when applied in combination with manipulations of the visual and the somatosensory inputs, would further augment the observations from in our previous work. <u>12,13</u>

Methods

Subjects

Twenty healthy young adults (ten males and ten females; age 24.05 \pm 5.34 years, height 1.70 \pm 0.09 m and mass, 69.7 \pm 15.3 kg) participated in this study. The average of their preferred walking speed (PWS) was 1.02 \pm 0.08 m/s. They were free from any neural or musculoskeletal problems and had no recent history of lower extremity injures that might have affected their gait. In addition, subjects were excluded from the study if they had a history of visual or vestibular deficits and scored above zero on the dizziness handicap inventory for a vestibular deficit. <u>28</u> Prior to the experiment, each subject gave informed consent as approved by University of Nebraska Medical Center Institutional Review Board.

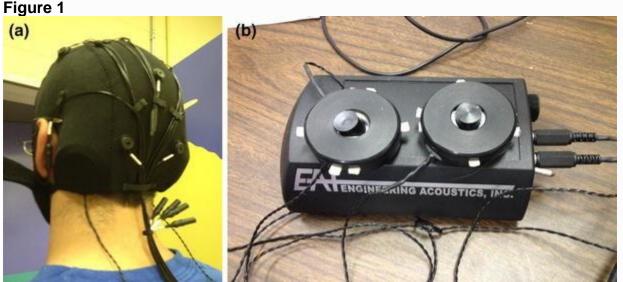
Instrumentation

The Locomotor Sensory Organization Test (LSOT) consists of two components: a virtual reality (VR) environment with a virtual corridor, and an instrumented treadmill (Bertec Corp., Columbus, OH, USA).<u>12,13</u> The LSOT contains six conditions similar to the Sensory Organization Test to manipulate sensory information during walking:

- 1. Normal walking condition: both the speed of the virtual corridor and the treadmill speed are matched with the PWS.
- 2. Reduced visual condition: no VR is presented, the treadmill speed is matched with the PWS, and the subjects wear vision-reduced goggles.
- 3. Perturbed visual condition: achieved by manipulating the optic flow speed. The speed of the virtual corridor is pseudo-randomly varied between 80 and 120% (restricted randomization between 80 and 120% in steps of 1%) of the selected PWS. Furthermore, these variations occur in pseudo-randomly assigned time intervals within 1–10 s (restricted randomization between 1 and 10 s in steps of 1 s)12,13,28,30,31 in order to reduce likelihood of adaptation of walking in the perturbed environment. The treadmill speed is matched with the PWS.
- 4. Perturbed somatosensory condition: achieved by manipulating the treadmill speed. The speed of the virtual corridor is matched with the PWS. The treadmill speed is varied between 80 and 120% of the PWS in pseudo-randomly assigned time intervals within 1–10 s. This experimental design is justified as walking speed is highly associated with the sensitivity of the somatosensory system and is crucial during stance-to-swing transition.<u>15,16</u>
- 5. Reduced visual and perturbed somatosensory condition: achieved by reducing vision and manipulating the treadmill speed. No VR is presented. The treadmill speed is varied between 80 and 120% of PWS in pseudo-randomly assigned time intervals within 1–10 s, and the subjects wear vision-reduced goggles.
- Perturbed visual and somatosensory condition: achieved by manipulating optic flow and treadmill speed. Both the speed of the virtual corridor and the treadmill speed are varied between 80 and 120% of the selected PWS in pseudorandomly assigned time intervals of 1–10 s duration. In this condition the velocity of the virtual corridor and treadmill are synchronized with a unitary gain relationship.

The MV used in the present study contained two electromechanical vibrotactile transducers (tactors; Engineering Acoustics, FL, USA.), that were placed on the mastoid processes bilaterally to perturb the vestibular feedback signals (Fig. 1). These tactors are designed for mounting within a seat or cushion, and can produce high force and displacement levels that allow the vibration to be easily felt even through layers of padding. The tactors require controllers and are designed for optimum vibrotactile efficiency at low frequencies (50-140 Hz). Their size is 4.8 cm in diameter and 1.9 cm in thickness. The frequency and amplitude of the stimulation are communicated wirelessly from a computer to the tactor controller unit, which transmitted the signals through cables to the tactors. In the present study, the frequency and amplitude of MV were set to 100 Hz and 17.5 db, respectively. These specific settings were selected based on extensive pilot and an evaluation of the available literature, 4,38 and were found to be the most effective to consistently induce changes in eye movement and sway during standing without producing any discomfort. A pulsed firing pattern with an active period duration of 0.3 s and a resting period duration of 0.6 s was used to prevent saturating the sensation of the vestibular system. Three conditions of MV were given to the participants: bilateral, unilateral or none (control). For unilateral stimulation, one side

was randomly selected for each subject at the beginning of the experiment and this side was used consistently for all of the unilateral trials.



(a) The tactors were secured in a cap and placed on the mastoid process on each side. (b) The tactor controller unit: for communication with the computer through Bluetooth and transmission of stimulus control signals to the tactors.

Subjects wore a safety harness attached to a LiteGait system (Mobility Research, AZ, USA) in order to increase safety whilst on the treadmill.

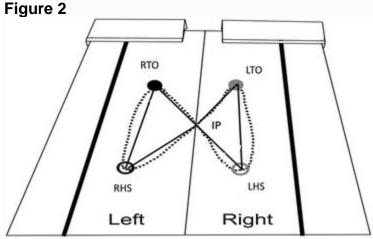
Procedures

Participants were required to complete 18 conditions (3 MV conditions by 6 LSOT conditions) on the same visit. The order of the presentation of the 18 conditions was randomized. Prior to the data collection, each subject walked for 5 min on the treadmill to determine their PWS. This commenced with the subject standing on the sides of the treadmill without touching the belts. The belt velocity was incremented from 0 to 0.8 m/s and the subject was asked to step onto the treadmill whilst holding the handrail. After the subject had started walking on the treadmill, experimenters asked the subject to evaluate the speed: "Is this walking speed comfortable, like walking around the grocery store?" The treadmill velocity was increased or decreased, based on subject directions. After a comfortable walking velocity had been attained, the subject walked continuously for 5 min. After the PWS had been determined, all subjects walked on the treadmill at their PWS for 2 min for each condition while data were captured. Between conditions, the subjects were asked to rest with closed eyes for 1 min. During this rest period, the subjects were asked if they have any feelings of discomfort. None of the subjects reported any such feelings of discomfort.

Data Reduction

The ground reaction force data acquired from the instrumented treadmill were low-pass filtered at 10 Hz (with a 4th order Butterworth filter). Then the netCOP trajectory is

identified from this data (Fig. 2).<u>36</u> Specifically, the netCOP is the point where the total sum of the pressure field acts on the body during walking. The total force vector acting at the netCOP is the value of the integrated vector pressure field.<u>36</u> For each gait cycle four specific netCOP points were identified: right heel strike (RHS), left heel strike (LHS), right toe-off (RTO), and left toe-off (LTO). These points identify two triangles that are created by the netCOP trajectory. One triangle has as vertices the LHS, LTO, and intersection point. The other has as vertices the RHS, RTO, and intersection point. The other has as vertices the netCOP area for each gait cycle (Fig. <u>2</u>). This netCOP area was used as an estimate of the postural sway during walking. In the current study, 85 gait cycles were used. This was the lowest number of gait cycles performed by these twenty participants in 2 min. The mean and the standard deviation for each subject were calculated by averaging the netCOP area over the 85 gait cycles. Then, the netCOP sway variability was calculated as the coefficient of variability.



The netCOP sway area was composed by two-triangle areas that are represented as the areas with solid lines. Five points was used to generate these two-triangle areas as following: intersection point (IP), right heel-strike (RHS), right toe-off (RTO), left heel-strike (LHS), left toe-off (LTO).

The temporal structure of sway variability was quantified using Sample Entropy (SampEn), calculated using a customized script in MatLab R2011a (Mathworks, Natick, MA). SampEn was preferred in the present study because was found to be more reliable that other Entropy algorithms (e.g., Approximate Entropy) for short data sets. <u>46</u> The SampEn was computed from the netCOP trajectory time series from the 2 min of available data. Data were downsampled from 12,000 to 1200 data points as we had observed little physiological signal above 10 Hz during our pilot studies. The SampEn algorithm is defined as the negative natural logarithm for conditional properties that a series of data points a certain distance apart, *m*, would repeat itself at m + 12.25 SampEn takes the logarithm of the sum of conditional probabilities. Given the time series g(n) = g(1), g(2), ..., g(N), where *N* is the total number of data points, a sequence of *m*-length vectors is formed. Vectors are considered alike if the tail and head of the vector are within the set tolerance level. The sum of the total number of like vectors is divided by m + 1 and defined as *A* or by N - m + 1 and defined as *B*. SampEn

is then calculated as $-\ln(A/B)$. A time series with similar distances between data points would result in a lower SampEn value while large differences result in greater SampEn value with no upper limit. Thus, a perfectly repeatable time series has a SampEn value = 0 and a perfectly random time series has a SampEn value converging toward infinity. In the current study, the following parameters were selected and used in the determination of SampEn values: (a) a pattern length (*m*) of 2, (b) and error tolerance (*r*) of 0.2.<u>46</u>

Statistical Analysis

Two-way fully repeated measures ANOVAs (3X6; 3 MV conditions by 6 LSOT conditions) were performed to determine statistical significance for the four dependent variables; (a) mean netCOP sway area, (b) coefficient of variation of the netCOP sway area, and (c) and (d) SampEn of the netCOP trajectory time series of the anterior—posterior and the medial–lateral directions. When significant main or interaction effects were determined, post hoc comparisons were performed using the Tukey method. Statistical analysis was completed in SPSS 18.0 (IBM Corporation, Armonk, NY) and the *a* value was set at 0.05.

Results

Mean netCOP Sway Area (Table 1)

A significant LSOT main effect (F = 2.88, p = 0.018) was found (Table <u>1</u>). However, the post hoc analysis did not reveal any significant differences between conditions due to the pairwise comparisons being adjusted for multiple comparisons. There was no significant MV main effect or interaction effect.

Table 1 Group condition means for netCOP sway area for 85 gait cycles per subject (m²).

Conditions	LSOT 1	LSOT 2	LSOT 3	LSOT 4	LSOT 5	LSOT 6
No MV	0.0493 ± 0.007	0.0493 ± 0.008	0.0498 ± 0.005	0.0515 ± 0.007	0.0495 ± 0.008	0.0503 ± 0.009
Unilateral MV	0.0497 ± 0.007	0.0486 ± 0.008	0.0482 ± 0.008	0.0511 ± 0.011	0.0480 ± 0.007	0.0506 ± 0.010
Bilateral MV	0.0493 ± 0.008	0.0488 ± 0.006	0.0486 ± 0.008	0.0497 ± 0.009	0.0468 ± 0.010	0.0508 ± 0.009

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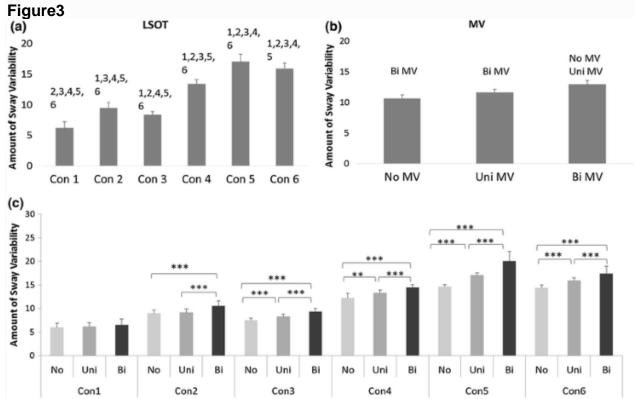
A significant main effect was found only for LSOT. No interaction effect was present. Post-hoc analysis using pairwise Tukey comparisons revealed no significant differences between conditions

LSOT Locomotor Sensory Organization Test, MV Mastoid Vibration

Amount of Sway Variability (As Measured by the Coefficient of Variation of the netCOP Sway Area; Fig. <u>3</u>)

A significant LSOT main effect (F = 1020.00, p < 0.0001) was found (Fig. <u>3</u>a). Post-hoc comparisons revealed that every LSOT condition was significantly different from all others. The largest value was present in condition 5, whilst the smallest was found for condition 1. In addition, a significant MV main effect (F = 200.58, p < 0.0001) was found

(Fig. <u>3</u>b). Post-hoc comparisons showed that the amount of sway variability was significantly larger in the bilateral MV condition than in the other two conditions. No differences were found between the unilateral MV and no MV conditions. A significant interaction was also identified between MV and LSOT (F = 12.03, p < 0.0001) (Fig. <u>3</u>c). Post-hoc comparisons showed that for normal unperturbed walking (LSOT Condition 1), MV did not produce any significant effect on the amount of netCOP sway variability. For LSOT condition 2, only bilateral MV significantly increased the amount of netCOP sway variability in comparison with no MV and unilateral MV. For the rest of the LSOT conditions, all possible comparisons were found to be significant with bilateral MV always producing the largest effect.

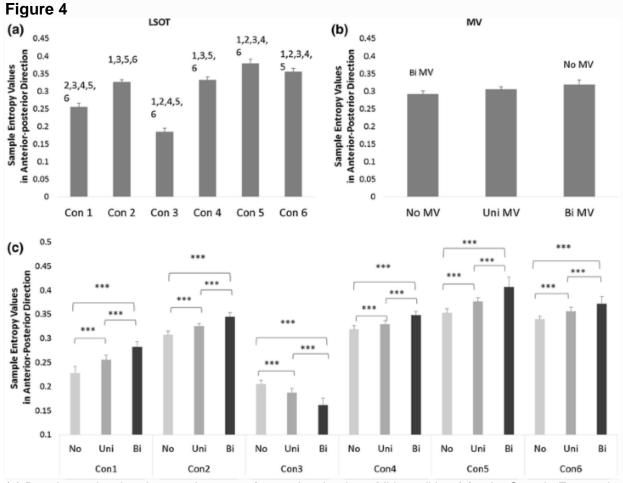


(a) Bar charts showing the margin means (averaging the three MV conditions) for the coefficient of variation of the six LSOT conditions. Error bars are standard deviations. The post hoc differences are indicated over the bars with the number of the condition with which differences were found. (b) Bar charts showing the margin means (averaging the six LSOT condition) of the coefficient of variation of the three MV conditions. Error bars are standard deviations. The post hoc differences are indicated over the bars with the three six LSOT condition. The post hoc differences are indicated over the bars with the type of the condition with which differences were found. (c) Bar charts for the group means (cell means in terms of the two-way ANOVA) for all conditions with brackets over the bars to identify significant differences between conditions. **<0.001; ***<0.0001.

Temporal Structure of Sway Variability in the AP Direction (As Measured by the SampEn of the netCOP Trajectory Time Series of the Anterior–Posterior Direction; Fig. <u>4</u>)

A significant LSOT main effect (F = 3122.01, p < 0.0001) was found for SampEn in the AP direction (Fig. <u>4</u>a). The post hoc tests revealed that all possible comparisons were significant with the exception of the comparison between conditions 2 and 4. Group

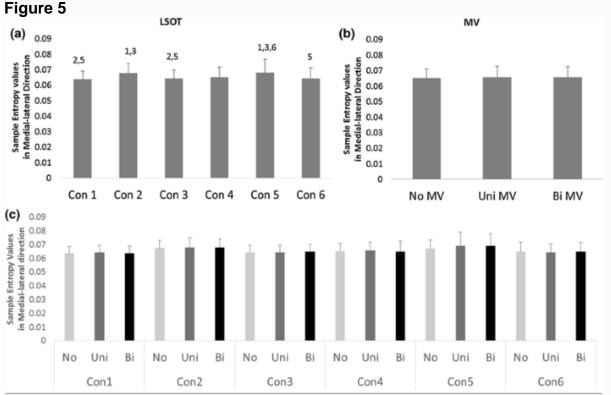
mean values were found to be at the lowest for Condition 3 and at the highest for Condition 5. A significant MV main effect (F = 275.24, p < 0.0001) was also found (Fig. <u>4</u>b). Post-hoc comparisons showed that bilateral MV condition produced significantly larger values than the no MV condition, while unilateral MV did not produce any differences with the other two conditions. A significant interaction was also found (F = 54.72, p < 0.0001) (Fig. <u>4</u>c). All post hoc comparisons were significantly larger values than the no MV condition, the unilateral MV produced significantly larger values than the no MV condition, while the bilateral MV produced significantly larger values than both the other two MV conditions. However, the opposite was the case for LSOT condition 3; bilateral MV produced the smallest value, while the no MV condition produced the largest.



(a) Bar charts showing the margin means (averaging the three MV conditions) for the Sample Entropy in the AP direction for the six LSOT conditions. Error bars are standard deviations. The post hoc differences are indicated over the bars with the number of the condition with which differences were found. (b) Bar charts showing the margin means (averaging the six LSOT condition) for the Sample Entropy in the AP direction for the three MV conditions. Error bars are standard deviations. The post hoc differences are indicated over the bars with the type of the condition with which differences were found. (c) Bar charts for the group means (cell means in terms of the two-way ANOVA) for all conditions with brackets over the bars to identify significant differences between conditions. **<0.01; ***<0.0001.

Temporal Structure of Sway Variability in the ML Direction (As Measured by the SampEn of the netCOP Trajectory Time Series of the Medial-Lateral Direction; Fig. 5)

A significant LSOT main effect (F = 9.85, p < 0.001) was found (Fig. 5a). Post hoc comparisons revealed that conditions 2 and 5 produced significantly larger values than conditions 1, 3, and 6. No significant differences were found between conditions 2 and 5. Condition 4 did not produce any significantly different results. No significant MV main effect or interaction was found (Figs. 5b and 5c).



(a) Bar charts showing the margin means (averaging the three MV conditions) for the Sample Entropy in the ML direction for the six LSOT conditions. Error bars are standard deviations. The post hoc differences are indicated over the bars with the number of the condition with which differences were found. (b) Bar charts showing the margin means (averaging the six LSOT condition) for the Sample Entropy in the ML direction for the three MV conditions. Error bars are standard deviations. No significant main effect was found. (c) Bar charts for the group means (cell means in terms of the two-way ANOVA) for all conditions. No significant interaction was found.

Discussion

We investigated how mastoid vibration could affect dynamic postural control in walking during simultaneous manipulation of the visual and the somatosensory systems. To accomplish this task we used three levels of MV (none, unilateral, and bilateral) and combined them with our LSOT paradigm.11,12 We used both amount and temporal structure measures of sway variability to investigate dynamic postural control.12,13 We hypothesized that the MV will affect both the amount and temporal structure measures of sway variability during walking and in combination with manipulations of the visual and the somatosensory inputs will further augment the results observed in our previous

research work. Our hypotheses were partially supported. MV produced significant increases for both measures of the amount and temporal structure of sway variability during walking. Regarding the temporal structure of sway variability, however, this was only the case for the AP direction but not the ML direction. Furthermore, for all conditions that involved visual and/or somatosensory manipulation, MV augmented the effect. This was the case regardless of whether MV was presented unilaterally or bilaterally. However, the bilateral MV stimulation produced larger effects than the unilateral. A notable exception to the above was LSOT condition 3 (the visual input is perturbed with no somatosensory manipulation being present) where MV resulted in decreased effects. Interestingly, MV affected only sway variability and not the mean sway area.

The overall lack of significant differences for the mean sway area could be due to the continuous adjustments the subjects made to step length and step width as they walked on the treadmill. Algorithmically, sway area highly depends on step length and step width. In our previous study we found that LSOT manipulations did not significantly affect step width. <u>12</u> Specifically, we observed an increasing trend from LSOT condition 1–6 with the largest difference between conditions to be about 0.09 m. On the other hand, step length significantly decreased and the largest difference between conditions was 0.12 m. Thus, it is possible that when we consider calculating sway area, the changes in step length and step width cancel each other out with our LSOT manipulations. In addition, MV produces no effect regarding this variable; however, MV did affect the variability of this variable.

Our results showed that MV further increased amount of sway variability during walking and this was the case for all LSOT conditions. Bilateral MV had a larger effect than unilateral MV, and the MV effect increased with increased difficulty of the LSOT condition presented (Fig. <u>3</u>c). Practically, these increases in variability reflect a significant positional drift towards the front and the back of the treadmill; as sensory input is affected, positional information during locomotion is compromised. These results lead us to suggest that MV, due to the affected vestibular input, causes confusion of the egocentric body-centered coordination system used during walking.<u>2,7</u> The increase in the amount of sway variability may be related to a response to correct the location of the netCOP to compensate for this confusion.

We found that manipulation of the vestibular input through MV does not produce a significant effect for amount of sway variability as we see in LSOT condition 1 (Fig. <u>3</u>c) unless combined with changes in another sensory input. Further, the size of the change produced by MV when just one other sensory input is manipulated (vision or somatosensory; LSOT conditions 2, 3, and 4) is quite similar. However, when both vision and somatosensory input are manipulated (LSOT conditions 5 and 6) and there is a greater reliance on vestibular input, MV produces much larger changes. Theoretically, sensory ambiguity could lead to greater uncertainty regarding necessary corrections compared to when a single modality is involved. When even less sensory input is available, the signal leads to a less accurate estimation of our position in space.<u>19,20</u> On the other hand, the larger changes could also be interpreted as an

effort to explore additional available space when less sensory input is present. In this fashion there is an effort to incorporate more solutions into the movement repertoire. Variability increases in this fashion are not viewed as a negative result but as positive result and scaling variability allows for the overall task to be accomplished within the available constraints.

Similar results to those observed for the amount of sway variability were produced in the AP direction for the temporal structure of sway variability, with few notable exceptions. MV further increased sample entropy values during walking. This was the case for five LSOT conditions. Overall, these changes in variability reflect significant alterations in the way positional drift towards the front and the back of the treadmill is temporally organized. Larger values of sample entropy reflect more uncertainty in the temporal structure and more irregular netCOP trajectory patterns. As sensory input is affected, positional information during locomotion becomes more convoluted and uncertainty is evident in the walking patterns as they evolve over time. With visual input perturbed but no somatosensory manipulation (LSOT condition 3), however, MV resulted in decreased effects demonstrated by more regular netCOP trajectories. This serendipitous finding should be validated via rigorous replication, however it is supported by Chien et al. who found that this LSOT condition produces more regular trajectories even when is compared with LSOT condition 1 (where no sensory input is manipulated).12,13 The question is then, why MV had an opposite effect in this condition in comparison with all others. It is likely that it is related to the manipulation used in this condition; perturbed visual input via a change in optic flow speed. Given that simply reducing vision as is the case in LSOT condition 2 does not have such an effect, we hypothesize that this finding is related to the intricate relationships between optic flow manipulation and MV through visual and vestibular input interactions. Manipulating optic flow affects the visual signal of self-motion,42 which could evoke the well-known vection sensations of self-motion9 and after-rotation when walking.26 This is combined here with MV that, as has been suggested, may affect space reference and thus locomotion.39 This hypothesis should be tested experimentally to further understand the mechanisms involved in the interaction of sensory inputs during locomotion.

Another interesting result that is different for the temporal structure of sway variability in comparison with the amount of sway variability, is that MV has an effect even when other sensory inputs are not being manipulated as is the case with LSOT Condition 1. Thus, it seems that MV, regardless of whether it is provided unilaterally or bilaterally, can affect the way the netCOP trajectories are organized in time producing more irregularity. This result suggests that vestibular input may be important for timing related movement decisions. Interestingly, unilateral local anesthesia of the upper dorsal cervical roots causes ataxia in humans, while ataxia and unsteadiness of gait characterize cervical vertigo.<u>7</u> Vestibular signals are frequency encoded around a central firing rate, but how they maintain a stable sense over time is not yet understood.<u>42</u> Our results support the notion that there is a closer relationship between vestibular inputs and timing of movements, regardless of whether we are dealing with unilateral or bilateral inputs.

Our results from both the amount and temporal structure of sway variability measures agree that bilateral MV produces a larger effect than the unilateral. Literature supports that bilateral and unilateral MV may produce different locomotor outcomes.5,6,27,33,35,41 Research has shown that continuous bilateral vibration of the dorsal neck muscles produces a reactive response in the sagittal plane and the AP direction resulting in a forward inclination of the body. 27, 33, 35 Ivanenko et al. suggested that, since the vestibular input is constant, such bilateral vibration could produce an illusion of the body's center of mass being located forward, "pressing for forward" propulsion of the body.27 On the other hand, unilateral mastoid vibration, as used in the present study, results in body turns to the side opposite to the vibration.5,6 In the context of treadmill walking, the lesser effects we observed with unilateral MV may be a result of the presence of external directional references provided by the experimental set up (e.g., fall harness, corridor, orientation of the moving belt) that allow the participant to recalibrate towards the anterior direction,41 thus countering the effect of the stimulation. This mechanism, similarly, may explain both the lack of a main effect of MV on the temporal structure of ML sway variability, and the modest differences between LSOT conditions in this direction. Conversely, the "pressing for forward" effect associated with bilateral MV would produce much larger results since AP is also the direction of motion.

In sum, our major conclusions were that MV produced significant increases for both measures of the amount and temporal structure of sway variability during walking. Regarding the temporal structure of sway variability, however, this was only the case for the AP direction but not the ML direction. Furthermore, for all conditions where visual and/or somatosensory manipulations were also introduced, MV augmented the effect regardless if was presented unilaterally or bilaterally. These conclusions should be tested if our experiments will be replicated with: (i) walking overground using technology that allows visual, somatosensory, and vestibular manipulations to be performed without the restrictions of the treadmill and safety harness; (ii) using a different direction of motion such as lateral stepping which will reverse the role of the AP and the ML directions for locomotion;44,45 (iii) using galvanic vestibular stimulation,23 dorsal neck muscles vibrations, 5,6 or changing head posture which affects balance and orientation responses.17,22,24 These experiments will allow us to eliminate alternative explanations of our results that were described above that arise from the proprioceptive contributions of the apparatus and the contribution of the mastoid vibration to vestibular inputs vs. other stimulations.

Abbreviations

MV: Mastoid vibration *LSOT:* Locomotor Sensory Organization Test *netCOP:* NET Center of Pressure *SampEn:* Sample Entropy

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