Human Proprioceptive Adaptations during States of Height-Induced Fear and Anxiety

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Abstract

Clinical and experimental research has demonstrated that the emotional experience of fear and anxiety impairs postural stability in humans. The current study investigated whether changes in fear and anxiety can also modulate spinal stretch reflexes and the gain of afferent inputs to the primary somatosensory cortex. To do so, two separate experiments were performed on two separate groups of participants while they stood under conditions of low and high postural threat. In Experiment #1, the proprioceptive system was probed using phasic mechanical stimulation of the Achilles tendon while simultaneously recording the ensuing tendon reflexes in the soleus muscle and cortical-evoked potentials over the somatosensory cortex during low and high threat conditions. In Experiment #2, phasic electrical stimulation of the tibial nerve was used to examine the effect of postural threat on somatosensory evoked potentials. Results from Experiment #1 demonstrated that soleus tendon reflex excitability was facilitated during states of height-induced fear and anxiety while the magnitude of the tendon-tap evoked cortical potential was not significantly different between threat conditions. Results from Experiment #2 demonstrated that the amplitudes of somatosensory evoked potentials were also unchanged between threat conditions. The results support the hypothesis that muscle spindle sensitivity in the triceps surae muscles may be facilitated when humans stand under conditions of elevated postural threat, although the presumed increase in spindle sensitivity does not result in higher afferent feedback gain at the level of the somatosensory cortex.

Keywords: Human, Posturography, Somatosensory Evoked Potential, Balance, Emotion
Introduction

Recent evidence has highlighted the potential for emotions, such as fear and anxiety, to influence human balance control. For example, young and older adults adopt different postural control strategies depending on the level of fear they experience when standing on the edge of an elevated surface height (Carpenter et al. 1999, 2001, 2006; Adkin et al. 2000, 2002; Davis et al. 2009; Huffman et al. 2009). However, the exact neural mechanisms that contribute to the observed changes in postural control remain unclear.

There is some evidence to suggest that changes in muscle spindle sensitivity may provide one mechanism through which fear and anxiety may influence postural control. For example, significant decreases in SOL H-reflex amplitude have been observed when individuals stand quietly on elevated surfaces (Sibley et al. 2007) or when they are engaged in a threatening task such as walking along a raised narrow beam (Llewellyn et al. 1990). This decrease in SOL H-reflex amplitude has been attributed to an increased tonic Ia discharge of muscle spindles which results in homo-synaptic post-activation depression of the SOL lower motor neuron pool (Sibley et al. 2007). Other evidence suggests that muscle spindles become more sensitive to stretch under threatening conditions; subjects standing on high compared to low surface heights demonstrated a significant increase amplitude of soleus tendon reflex (STR), without any accompanying changes in H-reflex amplitude (Horslen 2010). Similar observations of increased STR have been observed in seated individuals when presented with other sources of arousing stimuli (Bonnet et al. 1995; Both et al. 2005; Kamibayashi et al. 2009; Hjorstkov et al. 2005).

One explanation that has been offered for why spindle sensitivity may be increased in threatening or novel conditions is that it may “provide supraspinal areas with increased feedback gain and resolution” (Llwellyn et al. 1990). While there is current evidence indicating that later cortical potentials (ie N100) evoked by balance perturbations are increased under conditions of postural threat (Adkin et al. 2008; Sibley et al. 2010), the extent to which threat can
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influence the gain of afferent information initially received at the primary somatosensory cortex is currently unknown.

It is possible to determine whether an increased STR excitability also results in a larger magnitude afferent volley to the somatosensory region of the parietal cortex by simultaneously evoking STRs and cortical potentials (TEPs) via mechanical stimulations of SOL using tendon taps (Cohen et al. 1984; Frascarelli et al. 1993), although this technique has yet to be applied to standing individuals. Conversely, (SEPs) elicited by mild electrical stimulation of the tibial nerve allow for changes in the sensitivity of the somatosensory cortex to be measured independent of any changes in muscle spindle sensitivity (Gandevia and Burke 1984; Nelson et al. 2000; Staines et al. 2000; McIlroy et al. 2003).

Therefore, two experiments were performed to achieve the two aims of this study. Experiment #1 was designed to investigate whether elevated muscle spindle sensitivity during states of height-induced fear and anxiety results in facilitated spinal reflexes and a concomitant increase in the gain of afferent information delivered to the somatosensory cortex by comparing changes in STR excitability and TEP amplitude. Experiment #2 was designed to investigate the potential change in cortical sensitivity to incoming afferent information during states of height-induced fear and anxiety, independent of any changes in muscle spindle sensitivity, by comparing changes in SEP amplitude.

Based on previous observations (Horslen 2010; Bonnet et al. 1995; Hjorstkov et al. 2005; Kambayashi et al. 2009; Both et al. 2005; Llewellyn et al. 1990), it was hypothesized that there would be a significant increase in both TEP and STR amplitude when standing under conditions of elevated postural threat compared to when standing under conditions of low postural threat. Furthermore, it was hypothesized that there would be a null effect of threat manipulation on SEP amplitude reflecting no change in the sensitivity at the level of the somatosensory cortex to incoming afferent information, independent of any change in spindle sensitivity.
Methods

Participants

Two independent groups of participants volunteered for two separate experimental protocols in this study. In Experiment #1, 35 young healthy adults (17 female, mean ± SE; age 22.6 ± 0.6 years; height 173.9 ± 1.7cm; and weight 70.6 ± 1.8kg) volunteered to participate. In Experiment #2, 31 young healthy adults (16 male, mean ± SE; age 24.9 ± 0.8 years, height 174.5 ± 0.9cm; and weight 68.9 ± 0.4kg) volunteered to participate. All participants were recruited from the local undergraduate and graduate student community. Each participant completed a survey of their relevant medical history prior to testing. All participants were free of any relevant neurological, vestibular and/or orthopaedic conditions and were not taking any prescription medications that may have affected their balance performance during the study. Each participant provided written informed consent prior to testing. The University of British Columbia Clinical Research Ethics Board approved all experimental procedures.

Experiment #1: Mechanically-Evoked Somatosensory Potentials (TEP)

In Experiment #1, participants were separated into one of two groups: Free Standing or Braced Standing. During testing, those in the Free Standing group stood unsupported during all experimental procedures whereas those in the Braced Standing group stood in custom-made ankle braces designed to immobilize the ankle joint in the A-P direction (Figure 1). The ankle braces were used to prevent the characteristic posterior lean that occurs when participants stand under conditions of elevated postural threat (Davis et al., 2009). By maintaining a constant ankle angle in the Braced Standing group throughout the experiment (during both testing and rest periods), we attempted to control for any lean-associated changes in the length or tension of the muscles and tendon, the thixotropic state of the intrafusal muscle fibres, and tonic activation of the extrafusal muscle fibres, that could otherwise influence the amplitude of
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both the STR or TEP (Pierrot-Deseilligny and Mazevet, 2000; Proske et al. 1993; Gregory et al. 1998; Cohen and Starr, 1985).

Participants from both groups stood quietly at the edge of a hydraulic platform (2.13m x 1.52m surface area, M419-207B10H01D, Penta-lift, Canada) under two different conditions of postural threat; Low Threat and High Threat. During the Low Threat condition the hydraulic platform was resting at its lowest height (0.8m). Previous work has shown that standing 0.48m away from the edge of a platform at a height of 0.8m does not produce a postural effect that is significantly different from when standing on the ground (Carpenter et al. 2001). Therefore, a second support surface (0.61m x 1.52m) was placed in front of, and flush, with the front edge of the platform. During the High Threat condition, participants stood at the edge of the hydraulic lift after it was elevated to a height of 3.2m. While standing in each threat condition, participants were instructed to stand with their eyes open and fixated on a target placed at eye level 3.87m in front of them with their arms hanging freely and feet placed at the front edge of the platform with stance width equal to foot length. At all times, participants were securely harnessed to the ceiling for their safety in the event of a fall. The harness tether was kept slack and did not provide haptic cues or balance support during standing trials.

While standing in each threat condition, participants in both the Free Standing and Braced Standing groups received 2 blocks of 120 mild mechanical taps delivered to their right Achilles tendon for a total of 240 taps per threat condition. A 30s rest period was provided between blocks during which participants were instructed to lean backward slightly against the back of a chair that provided some postural support.

A computer driven magnetic linear motor (E2000-AT, LIN Mot, USA) was used as the tendon hammer to deliver mechanical taps to the Achilles tendon. The tendon hammer travelled a distance of 1cm and took 13ms to reach peak displacement. The distance between the tendon hammer and skin was measured at the beginning of each block of testing in each threat condition to ensure that a constant distance of 5mm was maintained between the tendon...
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hammer and the skin over the Achilles tendon throughout the experiment. Based on findings from initial pilot work, taps were applied to the Achilles tendon at an inter-stimulus interval of 1.2-1.7s to ensure that any postural perturbation caused by a given tendon tap and/or tendon reflex response fully subsided before a subsequent tap was delivered. The force of each tap was recorded with a dynamic force sensor (Isotron Dynamic Force Sensor and Conditioner, Endeveco, USA) mounted on the contact surface of the tendon hammer. The force of each tap was amplified 10x and sampled at 1000Hz (Spike 5, CED, UK). Clinical investigations of STR sensitivity have demonstrated that a tendon tap force ranging from 21-50N is adequate to elicit an STR in healthy young adults (Marshal and Little 2002). Therefore the force of each tap was kept within this range for all participants included in the study. In order to evoke an optimal STR, the tendon hammer contact location was adjusted in the vertical plane to ensure that the tap force for each participant was kept above sensory perception threshold and evoked a clear STR. The force of each tap was monitored on-line during testing to ensure that a consistent tap force was delivered between threat conditions and was recorded for off-line analysis.

STR and TEP Recording Procedure

In Experiment #1, electroencephalography (EEG) signals were recorded using the ANT WaveGuard cap system (ANT, Netherlands). The EEG electrodes were placed according to the international 10/20 system. The recording electrode was located along the sagital midline at the CZ position, and the reference electrode was located at Fpz’ position (2 cm caudal to Fpz). The common ground electrode was located along the sagital midline, between Cz and Fpz’ electrodes. The impedance between all three EEG electrodes was tested prior to and following the experiment (EZM5 impedance meter, Grass Instruments, USA) to ensure that the electrode impedance was less than 10Kohm during testing. All EEGs signals were amplified 20000x, sampled at 2048Hz (Porti7, ANT, Netherlands), band-pass filtered off-line between 1-100Hz.
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(MatLab, Mathworks, USA) and imported into a separate analytical software package (Spike5, CED, UK) for post-processing.

The STR was recorded by placing two surface Ag/AgCl electrodes ~4cm apart in a monopolar (belly-tendon) configuration on the right SOL. Monopolar electrodes are recommended for the purposes of recording muscle reflexes as they avoid signal cancellation problems associated with bipolar recordings (Hadoush et al. 2009). Background EMG activity was recorded by placing two surface electrodes ~2cm apart in a bipolar configuration on the right TA and SOL. The EMG electrodes shared a common ground with the EEG electrodes, which was located along the sagittal midline between Cz and Fpz’ in the WaveGuard cap. All EMG signals were amplified 2000x, sampled at 2048Hz (Porti7, ANT, Netherlands) and band-pass filtered (30-300Hz) off-line (MatLab, Mathworks, USA).

STR and TEP Analysis Procedure

During post-processing, each recording was inspected to remove any tendon taps that did not meet inclusion criteria for use in the calculation of the spike-triggered average of the STR and TEP. The inclusion criteria analysis was performed in a three step process. First, the ranges of the 240 tap forces delivered during each of the Low Threat and High Threat conditions were calculated for each participant. The condition with the narrowest range was used as the standard from which a mean ± 2SD threshold band was calculated. Second, individual taps that did not fall within this standard mean ± 2SD threshold band in either threat condition were removed from subsequent analysis. Prior to the experiment, subjects were asked to perform a series of voluntary eye blinks so that resulting blink artefacts could be identified within the EEG signal. In all cases the blink artefacts were very large, and easily discernable from any underlying cortical activity. Following the experiment, visual inspection of each EEG recording was then used to identify any blink artefacts, and any stimulation triggers that coincided with the timing of a blink artefact were deleted prior to the creation of the
ensemble average. Data from individual participants were only included in the subsequent
analysis if there were at least 100 stimuli that met these inclusion criteria for each threat
condition.

24 participants (10 female, mean ± SE; age 23.0 ± 0.7 years; height 175.3 ± 1.8cm; and
weight 70.9 ± 2.0kg) (12 in the Free standing group and 12 in the Braced Standing group)
passed the inclusion criteria analysis. For those 24 participants, spike-triggered averages were
calculated for the STR and TEP signal between 100ms prior to, and 300 ms after, the stimulus
onset (Spike 5, CED, UK). P1 and N1 peaks of the TEP waveform were identified and used to
calculate the times to peak, and the peak-peak amplitudes (Gandevia and Burke 1984; Cohen
et al. 1990). The peak-peak amplitude and latency of the STR were calculated from the spike-
triggered average of the STR from each threat condition. The P1-N1 component reflects early
cortical reception of the afferent volley delivered to the somatosensory cortex in response to
phasic mechanical and electrical stimulation of the tibial nerve (Gandevia and Burke 1984;
Cohen et al. 1990; Nelson et al., 2000). Background EMG activity in SOL and TA were
calculated as the mean level of activation of the full-wave rectified EMG signal 100ms prior to
stimulus onset.

Physiological and Psychosocial Estimates of Height-induced Fear and Anxiety

Electrodermal activity (EDA) of the non-dominant hand was used to provide an estimate of
the level of physiological arousal participants experienced during each threat condition.
Participants were fitted with disposable recoding electrodes on the thenar and hypothenar
eminences to measure their skin conductance (2502 Skin Conductance Unit, CED, UK) with a
range of 0-100 μmho. The skin conductance signal was collected at a sampling frequency of
1000Hz. EDA was averaged across the first 60s of each trial and subsequently averaged
across trials for each threat condition.
Before testing in each threat condition, participants were instructed to rate how confident they were that they would be able to maintain their balance and avoid a fall during the threat condition on a scale of 0% (no confidence) to 100% (complete confidence) (Adkin et al. 2002). Immediately following each standing trial, participants were instructed to rate how stable they felt during the trial on a scale of 0% (very unstable) to 100% (completely stable) and how fearful of falling they were during the trial again on a scale of 0% (completely unafraid) to 100% (extremely afraid) (Adkin et al. 2002). Participants also completed a 16-question survey of their perceived anxiety modified from Adkin et al. (2002). The scores from all 16 questions were summed to generate a total perceived anxiety score for each threat condition.

Statistical Analysis

All dependent variables were compared between standing conditions (Low Threat and High Threat) and between groups (Free Standing and Braced Standing) using 2x2 (condition x group) mixed design ANOVAs (SPSS, IBM, USA). Within-subjects effect sizes (Cohen’s d values) were calculated for each dependent variable as well. The criteria for statistical significance was set to p<0.05.

Experiment #2: Electrically Evoked Somatosensory Potentials (SEP)

In Experiment #2, participants stood under the same threat conditions described in Experiment #1: Low Threat and High Threat. While standing in each threat condition, participants received 2 blocks of 120 mild cutaneous electrical stimulations (S48 with SIU5 stimulus isolation unit and CCU1C constant current unit, Grass Instruments, USA) separated by a rest period of ~30s, for a total of 240 stimuli per threat condition (see details below). All participants in Experiment #2 stood unsupported; a braced standing group was not included. The rationale for not including a braced standing group was based on the fact that the location of SEP stimulation, the popliteal fossa, is proximal to the lower limb musculature. Therefore, the
ascending afferent volley evoked by the cutaneous electrical SEP stimulation would not have
been subject to any changes in muscle physiology or lower motor neuron pool excitability
associated with the threat-associated posterior COP shift.

Prior to the experiment, all participants performed a practice trial with the hydraulic lift
resting at its lowest height of 0.8m to remove potential first trial effects. The order of
presentation of Low and High Threat conditions was counter-balanced across participants to
minimize potential order effects. At the end of the experiment, participants performed a post-test
standing control trial in the Low Threat condition that was used to confirm the stability of the
SEP signals over time.

COP Measures

Ground reaction forces and moments were sampled (#K00407, Bertec, USA) at 100 Hz
(Power 1401, CED, UK) and low-pass filtered offline using a 5 Hz dual-pass Butterworth filter
(MatLab, Mathworks, USA) before calculating Centre of Pressure (COP) in the anterior-posterior
(A-P) direction. Mean position of the COP during each trial was calculated in the A-P direction
and subtracted from the COP signal. From this unbiased signal, the Root Mean Square (RMS)
and Mean Power Frequency (MPF) of COP displacement were calculated in the A-P direction.
Note that COP was not measured in Experiment #1. Pilot testing revealed that the braces
influenced the ability to measure normal COP displacements in the Braced Standing group, and
initial force from the hammer, and resulting STR, both represented dynamic postural
perturbations that confounded the COP signal in the Free-Standing group.

SEP Stimulation Procedure

SEPs were evoked by electrical stimulation of the right tibial nerve. The anode (10cm x
3cm, coal rubber pad, AMG Medical Inc, Canada) was placed just superior to the patella and
the Ag-AgCl cathode (0.25cm in diameter, Kendall, USA) was placed in the popliteal fossa. To
determine the optimal SEP stimulation intensity for each participant, a series of SOL H-reflexes were elicited via a sub-maximal 0.5ms square wave pulse to the tibial nerve. The SEP stimulus intensity was set to 40-60% of the intensity required to elicit a small M-wave, as this range of intensities has been shown to provide an optimal range for the SEP to fluctuate within and not become saturated (Gandevia and Burke 1984). However, this intensity was just above sensory perception threshold for each participant and was not sufficient to elicit a reliable SOL H-reflex or M-wave in our participants. Therefore, in order to ensure that the intensity of the stimulation to the tibial nerve did not change between standing conditions, a M-wave test pulse equal to 50% of M-max (DS5, Digitimer inc. UK) was evoked prior to and immediately following each 120s standing trial in a sub-group of ten participants. The peak to peak amplitude of each evoked M-wave was monitored in order to confirm the consistency of the stimulus over the duration of the testing period.

Somatosensory Evoked Potential Recording and Analysis Procedure

A different recording system from the one used in Experiment #1 was used to collect EEG in Experiment #2. Sintered Ag/AgCl scalp electrodes (EASYCAP, Germany) were placed along the sagittal midline at the Cz and Fpz’ (2cm caudal to Fpz) locations on the participant’s scalp according to the international 10/20 system for EEG recordings. A ground electrode was placed on the back of the participants’ neck (5cm x 5cm, coal rubber pad, AMG Medical Inc, Canada). The impedance between all three electrodes was tested before and after testing (Grass EZM5 impedance meter, Grass Instruments, USA) to ensure that the electrode impedance was kept at less than 10Kohm during testing. The EEG signal was amplified 20000x, sampled at 10000Hz and band-pass filtered between 1-1000Hz (Grass, P511 AC amplifier, USA). Each recording was visually inspected offline, to remove any stimuli that may have been contaminated by artifacts related to eye blinks or facial contractions. The remaining stimuli were used to create a spike-triggered average (Spike 5, CED, UK) of the SEP signal between 100ms prior to, and 150
ms after, the stimulus onset. The ensemble spike-triggered average was baseline corrected using the mean of the signal 100 ms prior to stimulus onset and band-pass filtered between 1-100Hz using customized software (LabVIEW, National Instruments, USA). P1 and N1 peaks of the SEP waveform were identified and used to calculate time to peaks, and the peak-peak amplitude of P1-N1 for each subject.

Two surface Ag/AgCl EMG electrodes were applied in a belly-tendon preparation on the right SOL muscle in order to record the SOL H-reflexes used for stimulus calibration. Two surface Ag/AgCl electrodes were placed ~2cm apart in a belly-belly preparation to collect background EMG activity in SOL. All EMG signals were amplified 2000x, sampled at a 1000Hz and band-pass filtered between 30-300Hz (P511 AC Amplifier, Grass Instruments, USA).

Physiological and Psychosocial Estimates of Height-Induced Fear and Anxiety

Both EDA and psychosocial estimates of height-induced fear and anxiety were recorded in the same manner described for Experiment #1.

Statistical Analysis

All dependent variables were compared between Low and High Threat conditions using paired-samples t-tests (SPSS, IBM, USA). Paired t-tests were also used to compare SEP amplitudes measured at the low height during the experiment, and the post-test control trial. Within-subjects effect sizes (Cohen’s d values) were calculated for each dependent variable as well. Correlation analysis was performed using a Pearson correlation on the changes between the high threat and low threat conditions for the physiological and posturographic variables. The criteria for statistical significance was set to p<0.05.

Results

Experiment #1: Mechanically Evoked Somatosensory Potentials (TEPs)
In the Low Threat condition, the TEP waveform was characterized by distinct P1-N1 component (Figure 2A). The mean time to peak for the P1 and N1 peaks occurred at 35.46 ± 0.92ms and 48.12 ± 0.96ms, respectively, following stimulus onset. The mean peak-peak amplitude of the P1-N1 was 4.80 ± 0.61μV. A similar TEP waveform was observed in the High Threat condition. There were no observable differences in the grand average waveforms between threat conditions with respect to the timing or amplitude of the P1-N1 component of the TEP waveform (Figure 2A).

The lack of observable differences in TEP timing and amplitude between Low and High Threat conditions was confirmed statistically. A mixed design ANOVA revealed no significant difference between threat conditions for the time to peak of the P1 \( (F(1,22)=2.311, p=0.143, \delta=0.13) \) and N1 \( (F(1,22)=0.335, p=0.568, \delta=0.07) \) when standing in the High Threat compared to Low Threat condition (Table 1). There was also no significant main effect of threat condition on the amplitude of the P1-N1 component \( (F(1,22)=0.904, p=0.352, \delta=0.07) \) of the TEP waveform (Table 1). However, there was a significant main effect of group on the peak-peak amplitude of the P1-N1 \( (F(1,22)=6.302, p=0.020, \delta=0.74) \) whereby larger P1-N1 amplitudes were observed in the Braced Standing group compared to the Free Standing group. There were no significant interaction effects between threat condition and group.

Despite very little change in the observed TEP (Figure 2B), there was a distinct increase in the STR peak-peak amplitude in the High Threat compared to the Low Threat condition (Figure 3C). This observation was confirmed statistically across participants. There was a significant main effect of threat condition on both the peak-peak amplitude \( (F(1,22)=4.430, p=0.047, \delta=0.12) \) and peak latency \( (F(1,22)=5.109, p=0.034, \delta=0.15) \) of the STR. The peak-peak amplitude of the STR was significantly larger and occurred significantly earlier when standing in the High Threat condition compared to the Low Threat condition, independent of whether participants were in the Free Standing or Braced Standing group (Figure 3AB). Furthermore, there was a significant main effect of threat condition on both the mean level of background SOL EMG
activity ($F_{(1,22)}=9.547$, $p=0.005$, $\delta=0.30$) and mean level of TA EMG activity ($F_{(1,22)}=9.895$, $p=0.005$, $\delta=0.48$). In both the Free Standing and Braced Standing group, there was a significant decrease in the mean level of background SOL activity and a significant increase in mean level of background TA activity when participants stood in the High Threat compared to the Low Threat condition (Figure 3C).

There was a significant main effect of threat condition on the level of confidence ($F_{(1,22)}=62.64$, $p<0.001$, $\delta=1.57$), perceived anxiety ($F_{(1,22)}=24.06$, $p<0.001$, $\delta=0.085$) and fear ($F_{(1,22)}=33.51$, $p<0.001$, $\delta=1.17$). As shown in Table 1, there was a significant increase in both self-reported fear and perceived anxiety and a significant decrease in self-reported confidence when standing in the High Threat condition compared to the Low Threat condition. There was also a significant main effect of height observed on EDA ($F_{(1,22)}=29.498$, $p<0.001$, $\delta=0.54$) whereby participants demonstrated higher EDA in the High Threat compared to the Low Threat condition (Table 1).

**Experiment #2: Electrically Evoked Somatosensory Potentials (SEPs)**

In the Low Threat condition, the mean SEP waveform was characterized by a P1 peak with a mean latency of 38.98 ± 1.44 ms following stimulus onset, followed by an N1 peak at 49.61 ± 1.78 ms (Figure 4A). In the High Threat condition the mean latency of the P1 peak was 38.98 ± 1.38 ms following stimulus onset, followed by an N1 peak at 50.55 ± 2.04 ms. There was no observable difference between threat conditions in the time to peak for any component of the SEP and there was no observed difference in the artefact amplitude (Figure 4A). Furthermore, there was no observable change in the amplitude of the P1-N1 component of the SEP between the Low Threat and High Threat conditions (Figure 4B).

The lack of observable differences in SEP amplitudes between Low and High Threat conditions was confirmed statistically (Figure 2B). Paired t-tests revealed no significant difference between the amplitude of the P1-N1 component ($t_{(30)}=-1.26$, $p=0.212$, $\delta=0.040$) of the
SEP waveform when standing in the High Threat compared to Low Threat condition (Table 1). Additionally, there was no significant difference between the time to peak of the P1 \((t_{30})=0.001, p=0.999, \delta<0.001\) and N1 \((t_{30})=-0.875, p=0.388, \delta=-0.089\) peaks when standing in the High Threat compared to Low Threat condition (Table 1). Furthermore, there was no significant difference between standing conditions in the peak-peak amplitude of the M-wave test pulse recorded in a sub-set of ten participants \((t_{9})=0.841, p=0.421, \delta=0.069\) confirming the stability of electrode placement during testing.

As shown in Figure 4C, there were significant changes in COP displacements observed between threat conditions. Participants’ mean A-P COP was shifted significantly further from the edge of the platform \((t_{30})=-7.867, p<0.001, \delta=0.836\), while MPF of A-P COP significantly increased \((t_{30})=-4.337, p<0.001, \delta=0.605\) and RMS of A-P COP significantly decreased \((t_{30})=4.622, p<0.001, \delta=-0.455\) when standing in the High compared to Low Threat condition (Table 1). Mean EDA increased significantly \((t_{30})=-4.794, p<0.001, \delta=0.923\) and tonic levels of SOL EMG activity did not significantly change \((t_{30})=0.785, p=0.439, \delta=0.134\) when standing in the High compared to Low Threat condition. There was also a significant increase in participants’ self-reported level of perceived anxiety \((t_{30})=-6.872, p<0.001, \delta=1.113\) and self-reported fear \((t_{30})=-6.916, p<0.001, \delta=1.261\) and a significant decrease in participants’ self-reported level of balance confidence \((t_{30})=5.538, p<0.001, \delta=1.024\) when standing in the High threat compared to Low Threat condition (Table 1).

Neither the change in P1-N1 amplitude or latency between the Low Threat and High Threat condition were correlated to the change observed in any of the physiological or psychological dependent variables. Furthermore, there was no significant correlation between the change in peak-peak amplitude of the M-wave test pulse and the change in either P1-N1 amplitude \((r^2 =0.027, p=0.651)\).

Discussion
The primary aims of this study were to determine whether muscle spindle sensitivity increases during states of height-induced fear and anxiety and whether this increase in spindle sensitivity results in higher afferent feedback gain at the level of the somatosensory cortex. The results from Experiment #1 demonstrated that STR excitability was facilitated and STR latency was reduced when participants stood in the High Threat condition compared to the Low Threat condition. These results are consistent with the facilitation of STR observed in subjects that were seated under conditions of increased arousal (Bonnet et al. 1995; Both et al. 2005; Kamibayashi et al. 2009; Hjorstkov et al. 2005). We would argue that our findings cannot be explained by posterior leaning, or consequent changes in length or tension of the muscle/tendon or intrafusal muscle fibres, since similar threat-related changes in STR were observed in both the Braced and Unbraced Standing group. Moreover, the observed increase in STR excitability in the High Threat condition cannot be explained by changes in background EMG activity because standing in the High Threat condition was associated with a significant decrease in SOL and a significant increase in TA EMG activity independent of bracing condition; and such a pattern of change in EMG background activity would lead to inhibition of the STR (Crone et al. 1987), as opposed to an increase in STR as currently observed. In this regard, the results suggest that not only is SOL STR excitability increased in High Threat conditions, but the increase is sufficiently large enough to overcome any inhibitory influence of the observed changes in background SOL and TA activity. Alternatively, it could be argued that the increase in STR amplitude under threatening conditions is caused by a decrease in pre-synaptic inhibition acting at the level of the spinal cord. However, evidence by Morita et al. (1998) have demonstrated that the T-reflex is less susceptible to influences of PSI, compared to the H-reflex amplitude, and there was no evidence for decreased H-reflex amplitudes in this or other studies that have controlled for postural leaning (Horslen 2010). Therefore, based on the current evidence, the most likely explanation for the observed increase in amplitude, and decrease in
latency, of STR, is an increase in muscle spindle sensitivity during conditions of increased threat.

Supraspinal Ia Afferent Signal Attenuation

Contrary to our original hypothesis, and that of Llewellyn et al. (1990), facilitation of STR excitability in the High Threat condition was not accompanied by an increase in the afferent signal recorded at the somatosensory cortex; no changes in TEP amplitude were observed between Low and High Threat conditions. Furthermore, the results from Experiment #2 clearly show that the sensitivity of the somatosensory cortex to incoming afferent information does not change during states of height-induced fear and anxiety as demonstrated by the null effect of threat condition on SEP amplitude. Instead, it appears that the heightened Ia volley associated with the increase in muscle spindle sensitivity and STR excitability, is somehow gated or dampened at the somatosensory cortex- or along the posterior-column medial-lemniscus pathway.

Gating of afferent sensory afferent information has been previously observed during different phases of gait or voluntary ankle movement (Abbruzzese et al. 1981; Rushton et al. 1981; Dietz et al. 1985; Duysens et al. 1995; Morita et al. 1998), perhaps as a means to facilitate voluntary motor control in such situations (Morita et al. 1998). Therefore, it is plausible that a similar gating mechanism may be activated in order to facilitate greater cortical control over posture during states of height-induced fear and anxiety (Huffman et al. 2009). However, if sensory gating or cortical inhibition was indeed responsible for the results of Experiment #1, an attenuation of the electrically-evoked SEP would have been expected to be observed in Experiment #2, which was not the case. The lack of observable changes in SEP amplitudes with postural threat is consistent with previous reports that early auditory-evoked potentials are not influenced by postural threat (Adkin et al. 2008). This is contrary to later cortical potentials, such
as the N100, that are significantly modulated by postural threat (Adkin et al. 2008; Sibley et al. 2010) or emotion (Bar-Haim et al. 2005; Eysenck et al. 1987).

One limitation of the current study is that TEPs and SEPs were evoked in different experimental populations. However, this protocol was deemed necessary to balance the needs of acquiring a sufficiently large number of TEP and SEP stimuli to generate reliable ensemble averages, while minimizing the time for subjects to habituate to the threat. Future studies could address this limitation by eliciting TEPs and SEPs from the same individuals. Likewise, further investigations of sensory gating and threat could incorporate recordings of evoked potentials from surface electrodes placed at different levels of the spine (Cameron et al. 2008; Murakami et al. 2008), or via indwelling thalamic electrodes in clinical populations (e.g. Katayama and Tsubokawa 1987; Klostermann et al. 2002) during states of height-induced fear and anxiety.

The Utility of Studying Changes in TEP Amplitude during Stance

Only two studies have examined TEPs in humans. Cohen et al. (1985) was the first to descriptively compare SEPs and TEPs evoked in participants lying prone. Similarly, the magnitudes of TEP responses to mechanical stretch of the Achilles tendon have been compared between the paretic and functional limbs of hemi-paretic patients while lying prone (Frascarelli et al. 1993).

Despite the observed null effect of height-induced fear and anxiety on TEP amplitude in the current study, the results provide the first evidence of TEPs recorded during stance. The mean peak-peak TEP P1-N1 amplitude (4.80 ±0.61 μV) and latency (35.46 ± 0.92 ms) measured during stance in the Low Threat condition is comparable to measures recorded from prone individuals by Cohen et al. (1984) (3.2 ± 3.1 μV amplitude; 27-37 ms latency) and Frascarelli et al. (1993) (3.67 ± 0.44 μV; no latency reported).

Of note, however was the observed main effect of bracing on TEP amplitude, with increased TEPs observed in the Braced compared to Unbraced Standing condition. One
possible explanation for the difference may be the additional cutaneous inputs provided by the braces. However, this explanation is unlikely, since prior evidence has shown that decreasing cutaneous sensitivity through cooling had no effect on TEP amplitude (Cohen and Starr, 1985). Alternatively, an increase in the sensitivity of the cortex may have been required to stand in the braces, as observed when subjects have been asked to control balance in other novel or challenging conditions (Staines et al. 2000). Since the SEP experiment did not involve a braced condition in this study, we cannot address this possibility, however the potential for TEPs to be modulated by such factors indicates that further research needs to be applied to this technique, within a standing model in particular.

**Potential Mechanisms and Significance of Increased Muscle Spindle Sensitivity**

If the increase in muscle spindle sensitivity does not serve to increase the gain of afferent input received at the somatosensory cortex as originally hypothesized (Llewellyn et al.1990), the question remains as to ‘how’ and ‘why’ the spindles are made more sensitive to stretch under conditions of increased threat.

As to ‘how’ muscle spindle sensitivity may be increased during conditions of increased threat, there are at least two possible mechanisms. One possible mechanism involves direct connections between the sympathetic nervous system and muscle spindles. There is evidence that feline muscle spindles receive direct innervation from the autonomic nervous system (Barker and Saito 1981) that facilitates increases in muscle spindle firing rates when activated (Hunt 1960). Likewise, performing mental arithmetic or a static handgrip contraction to directly activate the human sympathetic nervous system has been shown to facilitate stretch reflex excitability (Hjorstkov et al. 2005; Kambayashi et al. 2009). Therefore, the increased sympathetic response (indexed by elevated EDA) observed during the High Threat condition may have contributed to increased SOL muscle spindle sensitivity and facilitation of STR excitability in the current study. Although some studies have demonstrated that sympathetic
nervous system activation does not facilitate tonic muscle spindle firing rates in semi-intact animal preparations (Passatore et al. 1996; Hellström et al. 2005) or in humans (Macefield et al. 2003; Birznieks et al. 2008), it should be noted that in these studies, the muscles that were tested were not posturally relevant, or tested under a loaded condition.

Alternatively, the observed increase in spindle sensitivity and STR reflex facilitation observed may occur as a result of heightened gamma-motor drive to SOL muscle spindles (Prochazka et al. 1976, 1985, 1988; Ribot-Ciscar et al. 1986; Hospod et al. 2007). Evidence from cats has shown changes in Ia afferent firing patterns under difficult or threatening conditions (i.e. standing on a beam or ladder), that are consistent with patterns caused by activation of gamma static and dynamic neurons (Prochazka et al. 1985; 1988). While some studies have also provided evidence in support of alpha-gamma decoupling in humans (Ribot-Ciscar et al. 1986; Hospod et al. 2007), others have argued that selective modulation of γ-motor neuron activity in humans is unlikely (Gandevia et al. 1997). Therefore, more detailed experiments are clearly required to establish the potential influence of sympathetic drive and or alpha-gamma decoupling on muscle spindle sensitivity during stance.

At this point we can also only speculate as to ‘why’ the muscle spindle sensitivity may increase under conditions of increased threat. For example, it may be part of a more global fight/flight response, just as the sensitivity of other sensory receptors are known to increase with sympathetic activation (Hall, 2011). Alternatively, increased muscle spindle sensitivity under conditions of increased postural threat may serve a functional role in maintaining postural control. For example, the strategy of stiffening the ankle joint to maintain a tighter control of the COM during quiet stance has been frequently observed in conditions of increased postural threat (Carpenter et al. 1999; 2001), and would be potentially facilitated by increased gain of spinal stretch reflexes in the soleus muscles. How effective this may be, is questionable, particularly in light of some arguments that the small ankle rotations during quiet stance may be taken up mostly by the tendon, creating a ‘dead-zone’ in which the spindles are incapable of...
detecting the small changes involved in sway (Loram et al. 2009). More likely, the increased
muscle spindle sensitivity could be used to defend against unexpected postural perturbations to
balance, as a stretch of the soleus muscle would be countered by a larger, more rapid, plantar-
flexion torque to move the COM away from the edge of the high surface height. Although,
observations from unexpected platform rotations failed to demonstrate increases in
monosynaptic reflexes under conditions of increased threat, the observations of larger
automatic postural responses initiated within 100 ms of an unexpected platform rotation
(Carpenter et al. 2004), suggest that increased afferent gain may be used to modulate long-
latency reflex pathways. If this is the case, the current observations suggest that this afferent
input might be used to augment spinal-bulbar or spino-cerebellar reflex pathways, since no
changes were observed in either TEPs or SEPs recorded at the cortical level.

Conclusion

The current study provides further evidence that muscle spindle sensitivity in the triceps
surae muscles may be facilitated when humans stand under conditions of elevated postural
threat (Horslen, 2010; Sibley et al. 2007). The increase in spindle sensitivity observed in High
Threat conditions did not produce a concomitant increase in the magnitude of the afferent volley
received by the somatosensory cortex, nor was it accompanied by an observable change in the
cortical sensitivity to incoming afferent inputs generated by electrical stimulation of the nerve
above the spindle. These findings challenge the notion that muscle spindle sensitivity is
increased under difficult or threatening postural conditions to increase the gain of
somatosensory information received by the cortex (Llewellyn et al. 1990); instead, threat-
induced facilitation of the ascending afferent volley appears to be gated or inhibited at the level
of the brainstem, thalamus or cortex. Although the exact mechanism is currently unknown, the
results of the study provide new insight into how proprioception may contribute to threat-induced
changes in static and dynamic postural control and highlight the need for further study of the neurophysiological outcomes of fear and anxiety.

Acknowledgements

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Figure Legends

Figure 1:
The experimental set up used to support stance in the braced standing condition. Participants in the Braced Standing condition stood in custom-made ankle braces designed to attenuate postural sway in the anterior-posterior direction.

Figure 2:
TEP traces plotted for the group mean +/- SE (dashed lines) (A), and single representative subject (B). STR trace plotted from a single representative subject (C). Low threat conditions are shown in black lines, high threat conditions shown in grey lines. Dashed vertical line indicates the time of stimulus onset.

Figure 3:
Mean change (+/-SE bars) between the low threat and high threat conditions for SOL STR amplitude (A), SOL STR latency (B) and background EMG activity 100ms prior to stimulus onset for SOL (grey bar) and TA (white bar) (C). * indicates significant differences between threat conditions.

Figure 4:
SEP traces plotted for the group mean ± SE (dashed lines) (A) and single representative subject (B) Changes in the pattern of centre of pressure (COP) displacements from a single representative subject (C). Low threat conditions are shown in black lines, high threat conditions shown in grey lines. Dashed vertical line indicates the time of stimulus onset.

Table 1:
Mean change (+/-SEM) of all neurophysiological, physiological and psychosocial measures observed between the Low Threat and High Threat conditions. Significant main effects of threat condition are highlighted in bold text (p<0.05).
References


Horslen BC. Postural threat-induced modulation of stretch reflex pathways in static and dynamic postural control. [Online]. The University of British Columbia Faculty of Graduate studies. https://circle.ubc.ca/handle/2429/28643 [2010].

Fear and Proprioceptive Adaptation


Figure 1

- Safety Harness attached to the ceiling
- Computer controlled linear motor
- Adjustable mounting bracket
- Dynamic Force Sensor
- Custom-made ankle braces
- Hydraulic lift
Figure 3

A. Δ STR Peak-Peak Amplitude (mV)

B. Δ STR Peak Latency (ms)

C. Δ Background EMG (mV)

*
### Table 1

<table>
<thead>
<tr>
<th>Dependent Measure</th>
<th>Low Threat</th>
<th>High Threat</th>
<th>Low Threat</th>
<th>High Threat</th>
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<td><strong>Neurophysiological Measures</strong></td>
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<tr>
<td>P1-N1 Amplitude (µV)</td>
<td>4.80 ± 0.61</td>
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<td>P1 Latency (ms)</td>
<td>35.46 ± 0.92</td>
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<td>N1 Latency (ms)</td>
<td>48.12 ± 0.96</td>
<td>47.84 ± 0.80</td>
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<td><strong>Physiological Measures</strong></td>
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<td>Mean EDA (mOhm)</td>
<td>18.11 ± 1.91</td>
<td>24.72 ± 2.50</td>
<td>18.75 ± 1.41</td>
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<td>Mean SOL EMG (mV)</td>
<td>0.012 ± 0.002</td>
<td>0.009 ± 0.002</td>
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<td>Mean TA (mV)</td>
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<td>M-wave Amplitude (mV)</td>
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<td>2.06 ± 0.46</td>
<td>1.98 ± 0.43</td>
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<td><strong>Psychological Measures</strong></td>
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<td>Perceived Anxiety (sum score)</td>
<td>28.36 ± 1.75</td>
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<td>28.42 ± 1.49</td>
<td>54.68 ± 4.77</td>
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<td>Fear (%)</td>
<td>2.92 ± 1.27</td>
<td>24.79 ± 3.32</td>
<td>3.42 ± 1.32</td>
<td>37.84 ± 5.23</td>
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<td>Confidence (%)</td>
<td>96.87 ± 0.94</td>
<td>77.71 ± 2.50</td>
<td>95.35 ± 1.50</td>
<td>76.48 ± 3.75</td>
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<td><strong>Posturographic Measures</strong></td>
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<td>Mean A-P position (mm)</td>
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<td>-44.54 ± 2.72</td>
<td>-31.39 ± 2.70</td>
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<td>A-P MPF (Hz)</td>
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<td>0.226 ± 0.02</td>
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<td>A-P RMS (mm)</td>
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<td>4.68 ± 0.26</td>
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