Greater sensitivity of the vestibulosympathetic reflex in the upright posture in humans

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Sauder CL, Leonard TO, Ray CA. Greater sensitivity of the vestibulosympathetic reflex in the upright posture in humans. J Appl Physiol 105: 65–69, 2008. First published May 1, 2008; doi:10.1152/japplphysiol.90347.2008.—Otolith organs have been shown to activate the sympathetic nervous system in the prone position by head-down rotation (HDR) in humans. To date, otolithic stimulation by HDR has not been comprehensively studied in the upright posture. The purpose of the present study was to determine whether otolithic stimulation increases muscle sympathetic nerve activity (MSNA) in the upright posture. It was hypothesized that stimulation of the otolith organs would increase MSNA in the upright posture, despite increased baseline sympathetic activation due to unloading of the baroreceptors. MSNA, arterial blood pressure, heart rate, and degree of head rotation were measured during HDR in 18 normotensive, nonsmokers, nonobese, and unmedicated subjects.

Subjects

A total of 18 healthy volunteers (9 men and 9 women) [age: 23 ± 1 (SE) yr; height: 173 ± 3 cm; weight: 72 ± 4 kg] who were normotensive, nonsmokers, nonobese, and unmedicated were studied. Different subjects were used for study 1 and study 2. All subjects were studied for a total of 34 sessions. The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
received a physical examination by a physician before participation. Written, informed consent was obtained from all subjects after verbal explanation of the experimental protocol. The Institutional Review Board of The Pennsylvania State University College of Medicine approved the experiments.

Experimental Design

To determine the effect of posture on the vestibulosympathetic reflex, two studies were performed. Study 1 examined MSNA and cardiovascular responses to HDR in the prone and sitting positions, and study 2 examined responses to HDR in the prone and head-up tilt positions.

Experimental Protocols

Study 1. MSNA and cardiovascular responses to HDR were measured in the prone and sitting positions \((n = 11)\). The first trial examined responses to HDR in the prone position for 3 min. The second trial examined responses to HDR in the sitting position with the subject’s legs positioned horizontally with respect to the ground for 3 min. Both trials were performed on the same day and were preceded by a 3-min baseline period and followed by 3 min of recovery. The order of the trials was randomized.

The prone position trial began with the subject’s head in the baseline position. When in the baseline position, the head was upright with the neck extended as close to the vertical plane as possible and the chin supported. This position approximates the gravitational orientation of the head when an individual is in the upright posture \((24)\). For HDR, the head was maximally lowered in the vertical plane over the edge of the table. An investigator moved the head by supporting the forehead and chin, thus producing a passive head movement. Once the head became stationary, only afferent inputs from the otolith organs and not the semicircular canals are engaged. The sitting trial began with the subject’s head in the normal upright position. While in this position, the back of the head was supported by a headrest. The head was then maximally rotated forward in the vertical plane. MSNA, mean arterial blood pressure, heart rate, and degree of head rotation were measured continuously during both trials.

Study 2. MSNA and cardiovascular responses to HDR were measured in the prone and head-up tilt positions \((n = 7)\). The head-up tilt trial involved moving the subject from the supine position to 60° head-up tilt by an electric tilt table.

The head-up tilt trial began with a 3-min period in which the subject was lying in the supine position with the head flat against the table. Subjects were then tilted to a 60° angle for 15 min. This angle elicits \(\sim 87\%\) of the effects of gravity as one would experience during standing. Tilting permitted the measurement of MSNA, as actual standing would elicit electrical noise generated by the contracting leg muscles. Baseline data was collected in the upright posture with the subject’s head supported by the table. At minute 8 of head-up tilt, the subject’s head was passively lowered to the maximally flexed position by the investigator for 2 min. The subject’s head was then returned to the original position by the investigator until minute 14, and HDR was performed again for 1 min. The head was then returned to the original position by the investigator until the end of minute 15. The table was then returned to the horizontal position followed by a 3-min recovery period. The HDR trial in the prone position was as described in study 1. MSNA, mean arterial blood pressure, and heart rate were continuously measured during both trials.

Measurements

Multiplexer recordings of MSNA were obtained from a tungsten microelectrode inserted in the peroneal nerve behind or lateral to the knee, as previously described \((19)\). A reference electrode was placed subcutaneously 2–3 cm from the recording electrode. The criteria for an adequate MSNA signal included the following: 1) tapping of the muscles or tendons innervated by the nerve produced afferent mechanoreceptor discharges; 2) apnea produced an increase in sympathetic nerve activity; 3) stroking of the skin did not produce any afferent activity; and 4) sudden, unexpected arousal stimulus (shout or clap) did not produce any increases in sympathetic activity \((25)\). The nerve signal was amplified \((20,000–50,000\) times), fed through a band-pass filter with a bandwidth of 700–2,000 Hz, integrated by using a 0.1-s time constant \((University of Iowa Bioengineering, Iowa City, IA)\), and recorded digitally \((16SP PowerLab, ADInstruments, New Castle, Australia)\). The mean voltage neurogram was routed to a computer screen and a loudspeaker for monitoring during the study. Sympathetic recordings that demonstrated possible electrode site shifts, altered respiratory patterns \(e.g.\), breath holding, inspiratory gasp, and hyperventilation), or electromyographic artifact during experimental intervention were excluded from analysis. Microneurography was performed twice \(i.e.\), that is once in each position) on the same day for each study.

Angular rotation of the head was measured by a custom-built electrogoniometer. This allowed for the subject’s head position and degree of head rotation to be determined in all studies. The device was interfaced with an online computer and provided continuous tracking of any head movement and the degree of movement elicited by HDR. The degree of head rotation was \(\sim 106°\) in the prone position and \(40°\) in the sitting and head-up tilt positions.

Continuous measurements of arterial blood pressure and heart rate were made by finger plethysmography using a Finapres blood pressure monitor \((Ohmeda, Englewood, CO)\). Blood pressure and heart rate measurements were collected and analyzed offline. Electrocardiogram was obtained during all studies.

Respiration pattern was measured by a strain-gauge pneumotrace. This allowed for detection of inadvertent Valsalva maneuvers and apneas during the testing procedures.

Data Analysis

Sympathetic bursts were identified from individual inspection of the mean voltage neurograms and with computer assistance. Signal-to-noise ratio of 2:1 and a latency period of \(\sim 1.3\) s from the R wave of the ECG was required. MSNA was expressed as burst frequency \(\text{burst/min})\) and total MSNA \((i.e.\), sum of burst amplitude). The amplitude of the bursts was measured by a computer program \((Peaks; ADInstruments)\). Absolute changes from baseline are reported for burst frequency. Relative changes \%(from baseline are reported for total MSNA. For both the prone and the sitting trials, the 3 min of baseline were averaged together and reported as the baseline value for the respective trial. Additionally, the 3 min of HDR for each trial were averaged. During the head-up tilt trial, HDR responses from both trials were averaged together because they were not significantly different from each other. This included the 8th, 9th, and 14th minutes of head-up tilt. The minutes immediately preceding the two HDR trials were averaged together and used for baseline during head-up tilt. This included minutes 7 and 13 of head-up tilt. An index of the sensitivity of the vestibulosympathetic reflex was calculated by dividing MSNA changes elicited by HDR in each position by the change in head rotation (degrees). When calculating the sensitivity of the vestibulosympathetic reflex, the prone data for the two studies were averaged to give an overall mean. The data were analyzed using a one-between (posture) one-within (time) repeated-measures analysis of variance for both studies 1 and 2. Fisher’s protected least significant difference test was performed to determine whether responses to HDR were greater than baseline for each posture when there was a significant interaction \((posture \times time)\). One-way analysis of variance was performed on measures of vestibulosympathetic reflex sensitivity across the three postures. Least significant difference multiple-comparison post hoc tests were conducted when an overall significant effect was found. Significance was set at \(P < 0.05\). All data are presented as means \pm SE.
RESULTS

Study 1: Effect of Sitting on Responses to Head-Down Rotation

HDR elicited MSNA responses in both prone and sitting positions (Fig. 1). In the prone position, HDR increased MSNA burst frequency by $4 \pm 1$ bursts/min ($P < 0.01$) and total activity by $105 \pm 37\%$ ($P < 0.05$) (Fig. 1). In the sitting position, HDR increased MSNA burst frequency by $5 \pm 1$ bursts/min ($P < 0.01$) and total activity by $43 \pm 12\%$ ($P < 0.005$) (Fig. 1). Mean arterial blood pressure was increased by $\Delta 3 \pm 1$ mmHg ($P < 0.01$) and $\Delta 4 \pm 2$ mmHg ($P < 0.05$) for prone and sitting, respectively. Heart rate slightly increased in the prone position by $\Delta 2 \pm 1$ beats/min ($P < 0.05$) but did not change in the sitting position ($\Delta 1 \pm 1$ beats/min). Baseline measurements of MSNA, mean arterial blood pressure, and heart rate are reported in Table 1.

Study 2: Effect of Head-Up Tilt on Responses to Head-Down Rotation

HDR elicited increases in MSNA responses during prone and tilted positions (Fig. 2). In the prone position, HDR increased MSNA burst frequency by $3 \pm 1$ bursts/min ($P < 0.005$) and total activity by $36 \pm 13\%$ ($P < 0.05$) (Fig. 2). During head-up tilt, HDR increased MSNA burst frequency by $7 \pm 3$ bursts/min ($P < 0.05$) and total activity by $110 \pm 41\%$ ($P < 0.05$) (Fig. 2). Heart rate and mean arterial blood pressure were not significantly changed in either position. Baseline data for MSNA, mean arterial blood pressure, and heart rate are reported in Table 1.

DISCUSSION

There are two major findings from this study. First, activation of the otolith organs by HDR in the upright posture increases MSNA. Second, the sensitivity of the vestibulosym-
vestibulosympathetic reflex and posture. Unlike Watenpaugh et al., the present study examined MSNA and cardiovascular responses to otolith organ stimulation in the upright posture using the HDR model. HDR in the upright position clearly demonstrated increases in MSNA despite elevated baseline levels due to unloading of the baroreceptors.

There are several differences between the studies, which may account for these differences. First, Watenpaugh et al. (26) kept the head stationary once in the upright position. Thus it could not be determined whether MSNA could be augmented when the subject was upright, as with the present study in which HDR was performed after the subject was tilted upright. Second, the head was never positioned forward beyond the vertical axis as it is with HDR. Hume and Ray (11) demonstrated that head-down neck extension in the supine position elicited no changes in MSNA. Therefore, MSNA would not be expected to change in the head motion examined by Watenpaugh et al. However, we observed no changes in hemodynamic responses to a change in head position similar to that of Watenpaugh et al., but our findings do demonstrate marked increases in MSNA. This otolithic increase in MSNA in the upright posture has also been demonstrated by Kaufmann et al. (13). These investigators used off-vertical-axis rotation to stimulate the vestibulosympathetic reflex.

A second novel finding observed in the present study is that the sensitivity of the vestibulosympathetic reflex was greater in the upright posture. We observed that for a given degree of head rotation greater activation of MSNA was elicited. In a previous study, Hume and Ray (11) reported linear responses for MSNA during varying degrees of otolith organ activation using the HDR model in the prone position. We cannot determine whether this relation holds true in the upright posture because we only measured responses to one level of HDR. However, the results clearly indicate that MSNA responses to HDR are augmented relative to the degree of head rotation in the upright compared with the prone posture.

What mechanism is responsible for augmentation of the vestibulosympathetic reflex in the upright posture? The present study does not elucidate the mechanism. However, one possible mechanism is the central integration of the baroreflexes and the vestibulosympathetic reflex. Yates et al. (27, 29, 30) demonstrated that neurons regulating the vestibulosympathetic reflexes and baroreflexes synapse on the presympathetic neurons in the rostral ventrolateral medulla and on neurons in the nucleus tractus solitarius in the cat. These data suggest a possible integration between the two reflexes, enabling regulation of blood pressure during orthostasis. Ray (19) demonstrated an additive effect between the baroreflexes and the vestibulosympathetic reflex in humans during lower body negative pressure (−10 and −30 mmHg) and activation of the otolith organs during HDR. This study was performed in the prone position with low to moderate levels of lower body negative pressure to unload the baroreceptors. This finding was supported by Dyckman et al. (9), who used a steady-state infusion of sodium nitroprusside to unload the baroreceptors and then used HDR to stimulate the otolith organs. It is possible that greater unloading of the baroreceptors is needed to observe a facilitatory interaction between these reflexes.

A limitation of the present study is the inability to determine the contribution that each reflex (i.e., the baroreflexes and vestibulosympathetic reflex) has on sympathetic nerve activity.
when the subject is initially placed in the upright position prior to HDR. However, the present study successfully demonstrates that the vestibulosympathetic reflex can augment MSNA despite elevated basal levels of MSNA in the upright posture. It could be argued that another mechanism for increased MSNA during HDR in the upright posture is that HDR mechanically distorts the baroreceptors and therefore elicits an increase in MSNA. However, we have demonstrated previously that MSNA is not increased in subjects during HDR in the lateral decubitus position (20). This experimental design allows for maximal neck flexion and similar, possible distortion of the baroreceptors, whether it does occur with HDR, without activating the otolith organs. Therefore, it is believed that HDR does not mechanically activate the baroreceptors.

In summary, two significant and novel results were found in the present study. First, the vestibulosympathetic reflex can be activated in the upright posture via HDR, and second, the sensitivity of the vestibulosympathetic reflex is greater in the upright compared with the prone position. These findings support and further expand the concept that the vestibulosympathetic reflex is a robust reflex able to augment sympathetic nerve activity and contributes to the maintenance of blood pressure in the upright posture.

GRANTS

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