Cardiovascular responses to lower body negative pressure before and after 4 h of head-down bed rest and seated control in men and women


Faculty of Applied Health Sciences, University of Waterloo, Waterloo, Ontario, Canada; and Department of Medicine, University of Alberta, Edmonton, Canada

Submitted 4 June 2012; accepted in final form 6 September 2012

Edgell H, Grinberg A, Gagné N, Beavers KR, Hughson RL. Cardiovascular responses to lower body negative pressure before and after 4 h of head-down bed rest and seated control in men and women. J Appl Physiol 113: 1604–1612, 2012. First published September 13, 2012; doi:10.1152/japplphysiol.00670.2012—Cardiovascular deconditioning after a 4-h head-down bed rest (HDBR) might be a consequence of the time of day relative to pre-HDBR testing, or simply 4 h of confinement and inactivity rather than the posture change. Ten men and 11 women were studied during lower body negative pressure (LBNP) before and after 4-h HDBR and 4-h seated posture (SEAT) as a control for time of day and physical inactivity effects to test the hypotheses that cardiovascular deconditioning was a consequence of the HDBR posture, and that women would have a greater deconditioning response. Following HDBR, men and women had lower blood volume, higher heart rate with a greater increase during LBNP, a greater decrease of stroke volume during LBNP, lower central venous pressure, smaller inferior vena cava diameter, higher portal vein resistance index with a greater increase during LBNP, but lower forearm vascular resistance, lower norepinephrine, and lower renin. Women had lower vasopressin and men had higher vasopressin after HDBR, and women had lower pelvic impedance and men higher pelvic impedance. Following SEAT, brachial vascular resistance was reduced, thoracic impedance was elevated, the reduction of central venous pressure during LBNP was changed, women had higher angiotensin II whereas men had lower levels, and pelvic impedance increased in women and decreased in men. Cardiovascular deconditioning was greater after 4-h HDBR than after SEAT. Women and men had similar responses for most cardiovascular variables in the present study that tested the responses to LBNP after short-duration HDBR compared with a control condition.

cardiovascular deconditioning; orthostatic challenge; sex differences; regional blood flow

HEAD-DOWN BED REST (HDBR), as short as 4 h, is used as a simulation of microgravity exposure (6, 13). The body adapts quickly to this change in posture and reveals cardiovascular deconditioning that is characterized by elevations in heart rate (HR) and altered hormonal responses during application of lower body negative pressure (LBNP; i.e., lower baseline renin and angiotensin II which increased more in response to LBNP) (13) and by dramatic reduction in tolerance to an upright tilt (6). Thus acute alterations in the normal gravitational loading of the body appear to induce changes in the normal regulation of arterial blood pressure in the upright posture (13, 19, 35). But whether the changes during an orthostatic challenge after 4-h HDBR are an effect of the change in gravitational loading of the cardiovascular system or simply an effect of time of day and/or confinement has not been examined.

Inherent in 4-h HDBR studies is the comparison of responses at different times of day for pre- vs. post-HDBR measurements. Several studies have examined the cardiovascular and hormonal responses during the period of HDBR compared with equivalent durations of sitting, upright tilt, or ambulatory confinement as control experiments (5, 17, 21, 24, 31). However, only Pavy Le Traon and colleagues studied an orthostatic test after 4 days HDBR or an equivalent duration of ambulatory confinement finding that HDBR increased heart rate, reduced baroreflex sensitivity, decreased parasympathetic activity, and increased sympathetic activity; but, the same trends were seen, although with smaller magnitudes, after confinement (27). From this study, it is not clear how much of the HDBR effect might simply be a confinement effect.

Women have lower orthostatic tolerance than men probably as a consequence of greater reductions in cardiac filling (15). Women placed in HDBR were observed to have a smaller reduction in plasma volume than men, leading to speculation that women would be less affected on return to upright posture (14). However, in contrast to this proposal female astronauts are more likely than their male counterparts to experience orthostatic hypotension following spaceflight, possibly due to lower vascular resistance (20, 36). A recent theoretical consideration of sex differences in orthostatic tolerance after spaceflight postulated that the relatively larger mass of the lower body in women could account for the difference as women would have a relatively greater shift of fluid into the lower body, thus reducing cardiac filling (34).

The present study conducted a comparison of the cardiovascular and hormonal responses before and following 4 h of HDBR or 4 h of an upright seated control condition to test the hypothesis that the cardiovascular deconditioning was a consequence of the HDBR rather than being simply due to inactivity over the same time period. The study further tested the hypothesis that women would have characteristic changes reflecting a relative impairment of venous return compared with men.

MATERIALS AND METHODS

Ten men and eleven women between the ages of 25 and 32 yr participated in this study that was approved by the Office of Research Ethics at the University of Waterloo. Full written and verbal descriptions of the experiments were provided prior to obtaining written consent, and all subjects were informed of their right to withdraw at any time without penalty. The HDBR and the seated control experiments (SEAT) were completed in random order with a minimum of 48 h between test dates, but for women always within the range of days 8–11 of the menstrual cycle. Four of the women were not on any oral contraceptives (OC). Of the remaining seven women, three were...
taking cyclic types of OC (i.e., increasing levels of progesterone analog per week similar to normal physiology; Tricicyclin-lo, Ortho-McNeil Pharmaceutical, Raritan, NJ) and four were taking a noncyclic type of OC (i.e., the same level of progesterone analog every day; Alesse, Wyeth Canada, Montreal, QC, Canada). A recent study has shown that oral contraceptives do not modulate sympathetic neural and cardiovascular responses at rest or during an orthostatic challenge (7). Carter et al. also observed that in women taking OC there are no differences in the cardiovascular responses to LBNP in women taking cyclic (n = 9) vs. noncyclic OC (n = 3).

Subjects were requested to drink 5 ml/kg of water the night before testing and the morning before testing to provide a more consistent level of hydration. They reported to the laboratory at 7:00 am, 1 h after taking a light breakfast. Participants refrained from exhaustive exercise and alcohol for 24 h prior to testing and refrained from caffeinated beverages for 12 h prior to testing. Subject characteristics included body surface area (women 1.72 ± 0.07 m², men 1.94 ± 0.06 m²) and arm volume measured by water displacement (women 654.9 ± 69.6 ml, men 848.1 ± 50.7 ml).

Blood volume. On arrival in the laboratory, subjects voided their bladder, then a 20-gauge catheter (BD Insyte, BD Medical Systems, Sandy, UT, U.S.A.) was inserted into the antecubital vein of the right arm. Subjects sat upright in a comfortable chair for measurement of blood volume by a carbon monoxide (CO) rebreathing procedure described previously by Burge and Skinner (4) and used in this laboratory (13). Blood volume was used to normalize the diameter of the inferior vena cava.

LBNP and central venous pressure. LBNP was used as a model for orthostatic stress before and after the 4-h period of HDBR or SEAT. Central venous pressure (CVP) was measured throughout the LBNP test by the dependent arm technique (16). Individuals were placed supine in the LBNP box with the seal at the level of the iliac crest. The whole box was then tilted to the right to ensure filling of veins from the catheter back to the heart. The catheter in the antecubital vein was connected through a saline-filled line to a pressure transducer (Transine, Instrumentation for Medicine, Greenwich, CT) as described previously (5). All signals were recorded at 1,000 Hz with data acquisition hardware (PowerLab, ADInstruments, Arnheim, The Netherlands) and mean arterial pressure (MAP) was calculated as the average over each cardiac cycle. Heart rate (HR) was measured from the R-R interval of the electrocardiogram (Pilot 9200, Colin Medical Instruments, San Antonio, TX). Blood flow velocity from the aortic root and the brachial artery were recorded using 2-MHz and 4-MHz probes, respectively (model 500M, Multigon Industries, Yonkers, NY). Stroke volume (SV) was determined from the cross-sectional area of the aorta and the blood velocity of the aorta. Brachial arterial flow was determined from the cross-sectional area of the brachial artery and the blood velocity. Brachial vascular resistance was calculated as flow divided by MAP. SV was normalized to body surface area, and brachial vascular flow was normalized to arm volume. Biological impedance was measured in the thoracic region. Cross-sectional area of portal vein flow was calculated [portal flow (ml/min) = velocity × (π × radius²) × 60] then normalized to body surface area. Portal vein resistance index was determined by dividing mean arterial pressure by portal vein flow.

Cardiovascular measurements. Blood pressure was measured using finger-cuff photplessmography (Fenimeter; Finapres Medical Systems, Arnhem, The Netherlands) and mean arterial pressure (MAP) was calculated as the average over each cardiac cycle. Heart rate (HR) was measured from the R-R interval of the electrocardiogram (Pilot 9200, Colin Medical Instruments, San Antonio, TX). Blood flow velocity from the aortic root and the brachial artery were recorded using 2-MHz and 4-MHz probes, respectively (model 500M, Multigon Industries, Yonkers, NY). Stroke volume (SV) was determined from the cross-sectional area of the aorta and the blood velocity of the aorta. Brachial arterial flow was determined from the cross-sectional area of the brachial artery and the blood velocity. Brachial vascular resistance was calculated as flow divided by MAP. SV was normalized to body surface area, and brachial vascular flow was normalized to arm volume. Biological impedance was measured in the thoracic region. Cross-sectional area of portal vein flow was calculated [portal flow (ml/min) = velocity × (π × radius²) × 60] then normalized to body surface area. Portal vein resistance index was determined by dividing mean arterial pressure by portal vein flow.

Hormone analysis. Blood samples obtained during the LBNP testing were added to tubes containing appropriate anti-coagulants (norepinephrine: 25 μl/ml EDTA with glutathione; angiotensin II: 25 μl/ml EDTA with 20 μl/ml bestatin; vasopressin, renin: 25 μl/ml EDTA), then centrifuged at 2,500 rpm for 10 min at room temperature. Plasma renin was measured by radioimmunoassay (Active Renin IRMA, Diagnostic Systems Laboratories, Webster, TX), vasopressin was measured by radioimmunoassay (Vasopressin Direct RIA, ALPCO Diagnostics, Windham, NH), angiotensin II was measured by enzyme immunoassay (Angiotensin II Enzyme Immunoassay Kit, Société de Pharmacologie et d’Immunologie-BIO, Montigny Le Bretonnoux, France), and norepinephrine by HPLC with electrochemical detection (37).

Pelvic impedance. Pelvic impedance was measured using a Minnesota Impedance Cardiograph (Instrumentation for Medicine, Green-wich, CT). Pairs of electrocardiogram electrodes were placed at the xyphoid and iliac crest on both sides of the body (33). A constant alternating current of 4 mA at 100 kHz was passed through the pelvic region between the outer electrodes. The voltage was detected by the inner electrodes and was a product of the current and the regional impedance. All impedance measurements were normalized to the length of the respective leg.

Design and statistical analysis. The present study was designed to examine the effects of 4 h of SEAT or 4 h of HDBR on responses to...
LBNP in women during the low-hormone phase of the menstrual cycle and in men. The sex differences were compared by a three-way ANOVA with two repeated measures on main factors: women vs. men, level of LBNP, and pre- vs. post-SEAT or pre- vs. post-HDBR. Analysis of pelvic impedance, inferior vena cava compliance, and blood volume was done with two-way repeated measures ANOVA with one repeated factor for sex comparisons (HDBR or SEAT). Analysis was completed using SAS 9.1.3 analysis software. P values less than 0.05 are indicated as significant, and P values less than 0.10 are noted throughout.

RESULTS

Cardiac and vascular responses. During the SEAT control experiments, MAP was significantly decreased during LBNP in men and women (P = 0.009, Fig. 1A) and there was a trend for MAP to be lower in women after SEAT (P = 0.07, Fig. 1A). With HDBR, MAP was also reduced during LBNP (P = 0.003, Fig. 1B) and the significant interaction effect revealed a greater reduction of MAP in women than men (Sex \times LBNP, P = 0.001, Fig. 1B). Overall, there was no effect of HDBR on MAP (P = 0.436). In the SEAT experiments, heart rate was significantly elevated during LBNP (P < 0.0001, Fig. 1C) and there was a significantly greater increase in women (Sex \times LBNP, P = 0.046, Fig. 1C) but there was no difference due to SEAT alone (P = 0.514). In the HDBR experiments, HR increased due to LBNP (P < 0.0001) and HDBR (P < 0.0001). There was also a significantly greater increase in HR during LBNP following HDBR (HDBR \times LBNP, P < 0.0001, Fig. 1D). SV index was reduced by LBNP during the SEAT experiments (P < 0.0001, Fig. 1E) with no effect of SEAT (P = 0.973). In the HDBR experiments, SV index was reduced during LBNP (P < 0.0001, Fig. 1F) and the reduction was greater after HDBR (HDBR \times LBNP, P < 0.0001, Fig. 1F). Cardiac output index was also reduced by LBNP in both the SEAT and HDBR experiments (P < 0.0001, Fig. 2, A and B). The significant Sex \times LBNP in the SEAT (P = 0.009, Fig. 2A) reflects the slight elevation in cardiac output in men and reduction in women at baseline followed by a slightly greater decrease in men during LBNP. Total peripheral resistance was elevated during LBNP for both the SEAT and HDBR experiments (P < 0.0001, Fig. 2, C and D), and a significantly greater increase was observed in men only during the HDBR experiments (Sex \times LBNP; P = 0.009, Fig. 2D).

Blood volume and distribution. During the SEAT experiments, blood volume was unchanged from pre- to posttest in women (4,821 ± 463 vs. 4,810 ± 480 ml) and men (6,311 ± 805 vs. 6,244 ± 766 ml, P = 0.29) (main effect of SEAT). Blood volume was slightly but significantly reduced after 4 h HDBR [women: 4,836 ± 337 vs. 4,715 ± 347 ml; Men: 5,665 ± 697 vs. 5,486 ± 691 ml, P = 0.001 (main effect of HDBR)]. Serum osmolality was reduced in men after SEAT (282.0 ± 2.2 vs. 278.4 ± 2.0 mOsm/kg) but not in women (280.9 ± 1.8 vs. 280.7 ± 1.8 mOsm/kg; P = 0.044 Sex \times SEAT interaction). There were no differences in osmolality after HDBR for men or women.

During the SEAT experiments, portal vein resistance (PVR) increased in women and men during LBNP (P = 0.008; Fig. 3A) and tended to increase after SEAT (P = 0.10; Fig. 3A). During the HDBR experiments, PVR increased in women and men.
Fig. 2. Cardiac output index and total peripheral resistance index responses to LBNP in women (white bars) and men (gray bars) before (solid bars) and after (hatched bars) 4 h of seated control (SEAT) or 4 h of HDBR. *LBNP indicates a significant main effect of LBNP. *(Sex*LBNP) indicates a significant interaction effect of sex and LBNP.

during LBNP ($P < 0.0001$; Fig. 3B) and increased after HDBR ($P = 0.032$; Fig. 3B). Men had a greater increase of PVR with LBNP than women (Sex × LBNP, $P = 0.023$; Fig. 3B), and HDBR augmented the increase of PVR seen with LBNP in both women and men (HDBR × LBNP, $P = 0.039$; Fig. 3B). During the SEAT experiments, brachial vascular resistance (BVR) increased with LBNP in men and women ($P = 0.012$; Fig. 3C), and decreased after SEAT in men and women ($P = 0.013$; Fig. 3C). After HDBR, BVR decreased in men and women ($P = 0.048$; Fig. 3D). On both testing days BVR was higher in women ($P = 0.011$ and $P = 0.003$; Fig. 3, C and D).

Indicators of central blood volume were obtained from thoracic impedance, central venous pressure, and inferior vena cava diameter. Thoracic impedance provided an index of fluid shifts, where increases in impedance reflect movement of blood away from the heart. Women had higher thoracic impedance than men ($P < 0.001$ and $P = 0.002$; Fig. 4, A and B). LBNP increased impedance on both SEAT and HDBR testing days ($P < 0.0001$; Fig. 4, A and B) with a significantly greater increase in women (Sex × LBNP, $P < 0.0001$). Thoracic impedance was higher after SEAT ($P = 0.009$, Fig. 4A) but not HDBR ($P = 0.663$, Fig. 4B). There was a significant interaction effect between HDBR and LBNP which was likely due to the slight (yet not significant) reduction in thoracic impedance after HDBR (HDBR × LBNP, $P = 0.011$, Fig. 4B). Central venous pressure was reduced during LBNP ($P < 0.0001$, Fig. 4, C and D), and there was a significant interaction effect indicating that with the lower central venous pressure at baseline there were slightly smaller reductions during LBNP after SEAT (SEAT × LBNP, $P = 0.029$). HDBR caused a significant reduction in central venous pressure in women and men ($P = 0.001$). Significant interaction effects indicated greater reductions in central venous pressure during LBNP in women on the HDBR testing day (Sex × LBNP, $P = 0.035$), and again from the starting point of lower central venous pressure at baseline there were smaller reductions during LBNP after HDBR (HDBR × LBNP, $P = 0.007$). There was a tendency toward a three-way interaction effect ($P = 0.084$) indicating that women tended to have lower central venous pressure than men with LBNP after HDBR (Fig. 4D). The inferior vena cava diameter was not changed after SEAT ($P = 0.12$, Fig. 4E), but was reduced in men and women after HDBR ($P = 0.003$, Fig. 4F). A significant reduction in diameter was found during LBNP ($P < 0.0001$, Fig. 4, E and F). Estimated inferior vena cava compliance increased after SEAT in men but decreased in women (Sex × SEAT, $P = 0.043$; Table 1). Men had lower pelvic impedance than women ($P = 0.004$ and $P = 0.058$, Table 1, SEAT and HDBR testing days, respectively). On the SEAT experimental day, women had an increase in pelvic impedance after SEAT compared with a decrease in men (Sex × SEAT, $P = 0.043$; Table 1). On the HDBR experimental day, women had a decrease in pelvic impedance after HDBR compared with an increase in men (Sex × HDBR, $P = 0.026$; Table 1).

Vasoactive hormones. Lower body negative pressure (LBNP) increased the plasma concentration of norepinephrine on both testing days for women and men ($P < 0.0001$; Fig. 5, A and B). Norepinephrine was reduced after HDBR ($P = 0.030$; Fig. 5B). Plasma renin levels tended to increase during LBNP on the SEAT testing day ($P = 0.063$; Fig. 5C) and increased on the HDBR testing day ($P < 0.0001$; Fig. 5D). There was a reduction in plasma renin after HDBR ($P = 0.016$; Fig. 5D).
with a tendency for a greater decease in men (Sex × HDBR, \( P = 0.068 \); Fig. 5D). Men had higher renin levels than women on the HDBR testing day (\( P = 0.019 \); Fig. 5D). On the SEAT testing day, LBNP increased levels of angiotensin II in men and women (LBNP \( P = 0.038 \); Fig. 5E) and the SEAT position caused an increase in angiotensin II in women with a decrease in men (Sex × SEAT, \( P = 0.030 \); Fig. 5E). There were no effects of HDBR on angiotensin II (Fig. 5F).

Vasopressin was reduced during LBNP in men and women on the SEAT testing day (\( P = 0.015 \); Fig. 5G). Vasopressin tended to increase after SEAT (\( P = 0.057 \); Fig. 5G), particularly in women (Sex × SEAT, \( P = 0.095 \)) at the baseline time point (Sex × SEAT × LBNP, \( P = 0.093 \)). Vasopressin decreased in women but increased in men after HDBR (Sex × HDBR, \( P = 0.003 \); Fig. 5H).

**DISCUSSION**

This is the first study in men and women to investigate whether the changes in cardiovascular and hormonal responses to LBNP that follow 4-h HDBR were a consequence of the HDBR or whether similar responses might be observed after 4 h of confined sitting (SEAT). Consistent with the first hypothesis, the cardiovascular system did respond differently to 4-h HDBR than to 4-h SEAT. Specifically, during the post-HDBR application of LBNP the HR increased more and SV was reduced more. As well, indicators of changes in central blood volume (i.e., inferior vena cava diameter and thoracic impedance) and portal vein resistance were different during LBNP that followed HDBR but not after SEAT. Central venous pressure was reduced following HDBR but not SEAT, while the change in central venous pressure during LBNP was affected by both HDBR and SEAT.

This study also provided novel insight into the responses of men and women to LBNP before and after 4 h of HDBR and SEAT. Our primary results are generally consistent with the second hypothesis and imply that women have compromised mechanisms of venous return during orthostatic stress which could be due to splanchnic pooling. This could be a result of relatively elevated angiotensin II in women after SEAT, or the directionally opposite changes in vasopressin of women and men after HDBR might have contributed to the differences in blood volume distribution.

**Circadian rhythm and inactivity in men and women.** This study has shown that there were no changes in heart rate, stroke volume, mean arterial pressure, or portal vein resistance index during the LBNP testing due to sitting. This corresponds to previous investigations that have shown no changes in these variables (at least when comparing 8 am to 2 pm, the approximate testing times of this study) (1, 9, 30). However, Lathers et al. (24) did observe an increase in stroke volume and cardiac output in men in 42° of head-up tilt. This was likely an effect of the orthostatic stress. We conclude from these observations that both SEAT and circadian rhythm do not affect heart rate, stroke volume, mean arterial pressure, or portal vein resistance in men or women.

The responses of central venous pressure did not reveal a main effect of SEAT; however, central venous pressure was reduced during LBNP after SEAT in men and women. The small reductions in central venous pressure did not have an
impact on venous return as indicated by the absence of effect on heart rate and stroke volume.

Brachial vascular resistance was lower after sitting, suggesting an effect of circadian rhythm (26) or inactivity. Brachial vascular resistance was used as an index of limb vascular resistance as both arms and legs increase resistance to the same degree with LBNP (23). Therefore, these results are interpreted to mean that leg vascular resistance was also lower after sitting. This could have led to increased pooling of blood in the legs reducing venous return. The impedance results differed from whole body impedance which decreases during the day (32) but were consistent with a fluid shift to the lower part of the body with inactivity in the upright seated posture.

No changes in norepinephrine were expected (10, 21) or observed after 4 h of SEAT. Similarly, we did not observe changes in renin after sitting, but different findings have been reported concerning possible circadian effects. Previous investigations that maintained a supine posture throughout the day found that renin was lower in the afternoon (10, 22), but a previous study in which seated posture was maintained also found no change in plasma renin activity (21). Plasma angiotensin II was significantly different after SEAT between the sexes; women increased and men decreased angiotensin II. The cause of this is not known. There were no changes in vasopressin levels after SEAT even though the men had, consistent with a previous investigation of sitting (21), a slight reduction in serum osmolality which might be expected to cause lower vasopressin (3). Perhaps a longer period of sitting or inactivity might have affected vasopressin as another study found reduced concentration in the afternoon in men in supine posture (10).

Total blood volume was not different after 4 h of SEAT in either men or women, a finding consistent to a previous investigation of sitting (21). In the previous investigation a transient reduction in plasma volume probably contributed to the brief rise in hemoglobin that had vanished by 4 h; a time course study was not conducted in the current investigation. Other studies have noted that plasma volume and blood volume were slightly higher later in the day (8, 12), yet these changes were quite small (~3%). Overall, sitting for 4 h had only small effects on blood volume and volume regulatory hormones.

Table 1. Compliance of the inferior vena cava and pelvic impedance measurements of women and men during lower body negative pressure before and after 4-h head-down bed rest (HDBR) or seated control (SEAT)

<table>
<thead>
<tr>
<th></th>
<th>Pre</th>
<th>Post</th>
</tr>
</thead>
<tbody>
<tr>
<td>Compliance, cm/mmHg</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Women SEAT</td>
<td>0.12 ± 0.02</td>
<td>0.10 ± 0.03</td>
</tr>
<tr>
<td>Men</td>
<td>0.13 ± 0.02</td>
<td>0.15 ± 0.02††</td>
</tr>
<tr>
<td>Women HDBR</td>
<td>0.14 ± 0.02</td>
<td>0.11 ± 0.02</td>
</tr>
<tr>
<td>Men</td>
<td>0.15 ± 0.03</td>
<td>0.13 ± 0.02</td>
</tr>
<tr>
<td>Pelvic impedance, ohm/cm</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Women SEAT</td>
<td>2.42 ± 0.22</td>
<td>2.58 ± 0.20</td>
</tr>
<tr>
<td>Men</td>
<td>1.57 ± 0.17</td>
<td>1.30 ± 0.23‡‡</td>
</tr>
<tr>
<td>Women HDBR</td>
<td>2.91 ± 0.56</td>
<td>2.32 ± 0.34</td>
</tr>
<tr>
<td>Men*</td>
<td>1.56 ± 0.14</td>
<td>1.62 ± 0.18‡†</td>
</tr>
</tbody>
</table>

Values are means ± SE. *Significant main effect of sex. ††Significant Sex × SEAT interaction. ‡‡Significant Sex × HDBR interaction.

Fig. 4. Thoracic impedance, central venous pressure, and inferior vena cava diameter responses to LBNP in women (white bars) and in men (gray bars) before (solid bars) and after (hatched bars) 4 h of seated control (SEAT) or 4 h of HDBR. *Sex indicates a significant main effect of sex. *LBNP indicates a significant main effect of LBNP. *SEAT indicates a significant main effect of SEAT. *HDBR indicates a significant main effect of HDBR. *(Sex*LBNP) indicates a significant interaction effect of sex and LBNP. *(SEAT*LBNP) indicates a significant interaction effect of SEAT and LBNP. *(HDBR*LBNP) indicates a significant interaction effect of HDBR and LBNP.
HDBR in men and women. In both men and women, HDBR resulted in higher heart rate with an augmented increase during LBNP, a greater reduction of stroke volume during LBNP, a slightly lower thoracic impedance during LBNP and smaller inferior vena cava diameter. These results differ from those of Lathers et al. who found that 4 h of 5° HDBR resulted in lower cardiac output and higher total peripheral resistance (24). The results of Lathers et al. could have been a result of greater sympathetic activation due to eating closer to the 4-h time point than in the current study. In conjunction with the smaller inferior vena cava, central venous pressure was also reduced after HDBR as found in an earlier study of only men (13). It has been previously speculated that the lower central venous pressure could be a consequence of increased central vein compliance and/or changes in blood flow distribution leading to reduced venous return (5). We did not find evidence for this in our study of men and women as inferior vena cava diameter changed in proportion to pressure. There was, however, a small but significant reduction of blood volume that might have contributed to lower central blood volume and central venous pressure. The mechanism for the reduced blood volume in the current study does not appear to have been reduced fluid intake or increased urine volume as no differences were found in these variables between the SEAT and HDBR testing days (data not shown). Similar duration studies have previously shown no difference in urine output in head-up or head-down positions (5). It is possible that increased fluid extravasation into the lymphatic system (2) contributed to the reduction in plasma volume as a consequence of the increased portal vein resistance index after HDBR. Taken together, the findings from men and women of altered cardiac response with reduced venous pressure and volume point toward altered mechanisms involved in maintenance of venous return after HDBR and are consistent with findings after 4-h HDBR in men only (5, 13).

The finding of higher portal vein resistance index during LBNP after 4 h of HDBR in men and women suggests, in contrast to a previous investigation of men (13), that blood flow through the splanchnic region was reduced more after HDBR. In the previous study, participants were allowed to drink water closer to the afternoon LBNP test, but water...
drinking did not change portal vein flow in another study (28). In the current study, the Sex × LBNP interaction term suggested that women had a smaller change during LBNP than the men. It is possible that some between-study differences in methodology as well as the inclusion of women who seem to respond differently might have resulted in different findings between studies. Higher resistance in the splanchnic bed after HDBR should have facilitated venous return but this appears to have been counteracted by other vascular beds as overall total peripheral resistance was unchanged after HDBR. Brachial vascular resistance was lower after HDBR, as it was after SEAT. The change in brachial vascular resistance was similar to the observation after 56 days of HDBR of reduced leg vascular resistance in women (11). Thus, if both arm and leg vasculature had lower resistance to flow during LBNP after HDBR this could contribute to reduced central venous pressure and inferior vena cava diameter.

In an attempt to gain further confirmation of changes in portal vein resistance we employed pelvic impedance to show fluid shifts in the pelvic region. Higher pelvic impedance after HDBR in men was consistent with less blood in the pelvic region. However, in women decreased pelvic impedance after HDBR implied increased blood volume in the region which was not consistent with the elevated portal vein resistance index. In a mixed-sex population, 4 h of HDBR has been shown to have no significant effect on pelvic impedance (25). It is possible that women pooled blood in the pelvic organs after HDBR. Future investigations of the internal iliac artery might assist in resolving the apparent discrepancy about blood flow distribution in the pelvic region.

Hormonal responses to HDBR were clearly observed for plasma norepinephrine and renin concentrations but not for angiotensin II. The reductions in norepinephrine and renin were consistent with previous investigations (17, 21) and could reflect reduced peripheral vasoconstriction. Norepinephrine and renin increased during LBNP as a consequence of sympathetic nervous system activation; angiotensin II was not increased probably because of the short duration at each level of LBNP. Vasopressin was relatively higher in men and lower in women after HDBR even though there were no differences in osmolality.

Bearing in mind the effects of circadian rhythm and inactivity, the observed effects of 4 h of HDBR in men and women were 1) lower blood volume, 2) higher resting heart rate with a greater increase due to LBNP, 3) a greater decrease of stroke volume due to LBNP, 4) higher portal vein resistance index with a greater increase due to LBNP, 5) lower resting central venous pressure and smaller inferior vena cava diameter, and 6) lower norepinephrine and renin concentrations. There were different responses between men and women to HDBR, vasopressin was reduced in women and higher in men, and pelvic impedance was lower in women and higher in men. Overall, these results reveal that 4 h of HDBR has clear effects on the mechanisms that support venous return and cardiac output at rest and especially during a subsequent orthostatic stress.

Limitations. The design of the study focused on the effects of short-duration confinement in an upright seated posture or a HDBR position for 4 h on a subsequent test of LBNP applied in a supine posture. It was assumed that the effects of the 4-h posture would be carried over into the LBNP session. Because the subjects needed to eat and drink, subsequent measures could have been affected. Subjects were asked to eat a small breakfast at 6:00 am before coming to the lab to start our study at 7:00 am. In cirrhosis patients, Schiedermayer et al. (29) observed a postprandial increase in portal vein flow. Therefore, our portal vein data could have been affected by eating. However, the first portal vein images would have been taken at approximately 8:30 am, 2.5 h after eating. A lunch was provided within the first hour of the HDBR or SEAT condition in an attempt to match the effects on testing ~3 h later. Participants may have eaten later than asked in the morning, and if this is the case, morning portal vein flow may be elevated above a true baseline due to a postprandial increase of flow.

There may have been an impact of moving the subjects from the chair to the LBNP test, but this was expected to be minimal as transitions required only 5–10 s.

Finally, before starting the afternoon LBNP test, participants were strongly encouraged to urinate; however, some participants elected to wait until after the LBNP test when they could use a washroom to collect the urine sample. This would have affected the pelvic impedance, but it was a balanced effect with men and women electing this option ~25% of the time.

Significance and perspectives. The results of the current study reveal that 4-h HDBR caused changes in cardiovascular and hormonal responses that were not simply a consequence of physical inactivity or time of day comparisons. The importance of controls has been investigated previously during the postural alteration (5, 17, 21) but responses to an orthostatic challenge have not been assessed after HDBR and equivalent duration of seated control. In contrast, factors involved in regulation of heart rate, stroke volume, venous pressure and volume, peripheral and splanchnic vasoconstriction, and hormonal regulation were all altered directly by HDBR. These results are important as they reveal the impact of short-duration alterations in gravitational loading on cardiovascular control in both men and women. That is, periods of HDBR, and possibly supine rest, might impact blood pressure regulation on transition to an upright posture. Such an effect has been proposed for the first rise of the day in acutely ill older persons (18). Knowledge gained from the HDBR model can be applied to other populations that experience orthostatic symptoms on assuming an upright posture.

ACKNOWLEDGMENTS

We thank Danielle Greaves and Jing Ouyang for excellent technical assistance.

GRANTS

This research was supported in part by grants from the Natural Sciences and Engineering Research Council of Canada (RGPIN6473-07), and the Canadian Space Agency (SSEP-2006) to R. L. Hughson. H. Edgell was supported by a Natural Sciences and Engineering Research Council of Canada graduate scholarship and an Ontario Graduate Scholarship. K. Beavers was supported by a Natural Sciences and Engineering Research Council of Canada graduate scholarship.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS

script; H.E. and R.L.H. edited and revised manuscript; H.E., A.G., N.G., K.R.B., and R.L.H. approved final version of manuscript.

REFERENCES


