Nervous System

Sex Differences in Sympathetic Neural-Hemodynamic Balance
Implications for Human Blood Pressure Regulation

Emma C. Hart, Nisha Charkoudian, B. Gunnar Wallin, Timothy B. Curry, John H. Eisenach, Michael J. Joyner

Abstract—Among young normotensive men, a reciprocal balance between cardiac output and sympathetic nerve activity is important in the regulation of arterial pressure. In young women, the balance among cardiac output, peripheral resistance, and sympathetic nerve activity is unknown. Consequently, the aim of this study was to examine the relationship of cardiac output and total peripheral resistance to muscle sympathetic nerve activity in young women. Multiunit peroneal recordings of muscle sympathetic nerve activity were obtained in 17 women (mean±SEM: age 24±3 years) and 21 men (mean±SEM: age 25±5 years). Mean resting muscle sympathetic nerve activity was lower in women compared with men (19±3 versus 25±1 bursts minute⁻¹; P<0.05), as was mean arterial pressure (89±1 versus 94±2 mm Hg; P<0.05). Mean arterial pressure was not related to muscle sympathetic nerve activity in men (P=0.80) or women (P=0.62). There was a positive relationship between total peripheral resistance and muscle sympathetic nerve activity (r=0.62; P<0.05) and an inverse relationship between cardiac output and muscle sympathetic nerve activity (r=−0.69; P<0.05) in men. Unexpectedly, muscle sympathetic nerve activity had no relationship to either total peripheral resistance (r=−0.27; P>0.05) or cardiac output (r=0.23; P>0.05) in women. Our results demonstrate that men and women rely on different integrated physiological mechanisms to maintain a normal arterial pressure despite widely varying sympathetic nerve activity among individuals. These findings may have important implications for understanding how hypertension and other disorders of blood pressure regulation occur in men and women.

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Key Words: sympathetic nerve activity ■ blood pressure regulation ■ sex ■ peripheral resistance

Interindividual differences in central and peripheral hemodynamics have become increasingly important to our understanding of arterial pressure regulation.¹⁻⁴ In young healthy men, total peripheral resistance (TPR) is positively related to muscle sympathetic nerve activity (MSNA), suggesting that MSNA is a good index of net whole body vasoconstrictor tone. However, men with higher levels of MSNA and TPR do not necessarily have higher resting arterial pressure.⁵ In previous studies, young men with high MSNA had a lower cardiac output and less α-adrenergic receptor vasoconstrictor responsiveness, reducing the net effect of high MSNA on arterial pressure.⁵

In young women, blood pressure is typically lower than that observed in men of the same age.⁶⁻⁷ Furthermore, the incidence of orthostatic hypotension is greater in women than in men,⁸ and women have lower tonic autonomic support of baseline arterial pressure.⁹ Women also exhibit blunted vasoconstrictor responses to α-adrenergic stimulation,¹⁰ which may be related to the vasodilator effect of estrogen.¹¹⁻¹⁶ These observations suggest that the contribution of vasoconstrictor tone to arterial pressure regulation differs in women and men. We, therefore, sought to compare the relationships among MSNA, cardiac output, and TPR in women and men. We hypothesized that, because young women have less autonomic support of blood pressure and decreased basal α-adrenergic vascular responsiveness compared with men, there would be a blunted relationship between MSNA and TPR and between MSNA and cardiac output in women compared with men.

Methods

Subjects

After the protocol for the study was approved by the institutional review board of the Mayo Clinic, 21 men and 17 women gave their written informed consent to participate in this study, and the study was completed in accordance with the Declaration of Helsinki. The subjects were recreationally physically active nonsmokers with no history of cardiovascular or other chronic diseases. The participant demographics are outlined in Table 1. Participants were asked to not consume anything except small volumes of water within 2 hour of the experiment and were asked to

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Table 1. Demographic Variables in Men (n=21) and Women (n=17)

<table>
<thead>
<tr>
<th>Demographics</th>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>25±1</td>
<td>24±1</td>
</tr>
<tr>
<td>Body mass, kg</td>
<td>80±2</td>
<td>62±2*</td>
</tr>
<tr>
<td>Height, cm</td>
<td>180±2</td>
<td>168±1*</td>
</tr>
<tr>
<td>BSA, m²</td>
<td>2.0±0.3</td>
<td>1.7±0.03*</td>
</tr>
<tr>
<td>BMI, kg m⁻²</td>
<td>24.6±0.6</td>
<td>21.9±0.9*</td>
</tr>
</tbody>
</table>

BSA indicates body surface area; BMI, body mass index. Data are means±SEMs.

*Data are different from men (P<0.05).

abstain from caffeine or alcohol consumption 24 hours before the study. To minimize the effects or reproductive hormones on autonomic control or cardiovascular function, all of the women were studied in the early follicular phase of the menstrual cycle or in the low hormone phase of oral contraceptive use.¹⁷

Measurements

All of the studies were performed in a clinical research unit laboratory at the Mayo Clinic, where ambient temperature was controlled between 22°C and 24°C. On arrival to the laboratory, subjects rested in the supine position during instrumentation. After local anesthesia with 2% lidocaine, a 5-cm, 20-gauge arterial catheter was placed in the brachial artery of the nondominant arm, using an aseptic technique. The catheter was connected to a pressure transducer, which was positioned at the level of the heart and interfaced with a personal computer to monitor arterial pressure. A 3-lead ECG was used for continuous recordings of heart rate.

Multunit MSNA was measured from the right peroneal nerve at the fibular head using insulated tungsten microelectrodes. A muscle sympathetic fascicle was identified when taps on the muscle belly or passive muscle stretch evoked mechanoreceptive impulses, and no afferent neural response was evoked by skin stimuli.¹⁸,¹⁹ The recorded signal was amplified 80 000-fold, band pass filtered (700 to 2000 Hz), rectified, and integrated (resistance-capacitance integrator circuit time constant 0.1 seconds) by a nerve traffic analyzer. Cardiac output was measured using an open-circuit acetylene uptake technique, as described previously.²⁰ The cardiac output was estimated immediately after each maneuver using the calculation method described previously.²¹,²² This technique has been validated against direct Fick measurements of cardiac outputs over a range of values and has a variability of ~7% at rest.²⁰ The instrumentation period included a practice cardiac output measurement to familiarize the subject with the procedure.

Protocol

After placement of the arterial catheter, participants were asked to rest supine during instrumentation for microneurography. Once a satisfactory site for measurement of MSNA was located, 15 minutes of baseline data were recorded with the subject resting quietly. Subsequently, duplicate measurements of cardiac output were obtained.

Data Analysis

Data were sampled at 240 Hz and stored on a personal computer for offline analysis. Mean arterial pressure (MAP) was calculated as the time integral over the pressure pulse. Systolic and diastolic blood pressures, MAP, heart rate, and MSNA were taken at the 4-minute period immediately preceding the first cardiac output measurement. Cardiac output is reported as the mean of the 2 measurements for each individual. Stroke volume was calculated as the cardiac output/heart rate, and TPR was calculated as MAP/cardiac output.

Sympathetic bursts in the integrated neurogram were identified using a custom-manufactured automated analysis program²³; burst identification was then corrected via visual inspection by a single observer. The program then compensated for baroreflex latency and associated each sympathetic burst with the appropriate cardiac cycle.

Statistical Analyses

Group data are expressed as means±SEMs. Differences between cardiovascular variables and MSNA in men and women were evaluated using 2-tailed independent t test. To assess the relationship between MSNA and cardiovascular variables, linear regression analysis was performed and Pearson’s correlation coefficients calculated. The critical α-level was set at 0.05, and data were analyzed using SigmaStat software (version 2.03, SPSS Inc).

Results

Group-Averaged Data for Cardiovascular and Neural Variables in Men and Women

The young women had a lower baseline systolic blood pressure, diastolic blood pressure, MAP, stroke volume, and cardiac output compared with men (Table 2; P<0.05), whereas heart rate was similar between sexes. Women had a lower body mass and body surface area compared with men (Table 1; P<0.05). Therefore, we scaled cardiac output for body surface area, which eliminated the difference in cardiac output between men and women (Table 2; P>0.05). The absolute TPR was higher in women compared with men (P<0.05); however, when cardiac output scaled for body surface was used to calculate TPR, the difference between men and women disappeared (P<0.05). Baseline MSNA was lower in women compared with their male counterparts when MSNA was expressed as burst frequency (bursts minute⁻¹) and incidence (bursts 100 heartbeats⁻¹; P<0.05).

Interindividual Relationships Between Cardiovascular and Neural Variables in Men and Women

Relationships between MSNA and cardiovascular variables are shown in Figure 1 with MSNA expressed as burst frequency. Data for burst incidence showed similar trends to

Table 2. Cardiovascular and Neural Variables in Men (n=21) and Women (n=17)

<table>
<thead>
<tr>
<th>Cardiovascular/Neural Variables</th>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td>HR, beats min⁻¹</td>
<td>59±2</td>
<td>63±2</td>
</tr>
<tr>
<td>SBP, mm Hg</td>
<td>136±3</td>
<td>126±1*</td>
</tr>
<tr>
<td>DBP, mm Hg</td>
<td>73±1</td>
<td>70±1*</td>
</tr>
<tr>
<td>MAP, mm Hg</td>
<td>94±2</td>
<td>89±1*</td>
</tr>
<tr>
<td>SV, mL</td>
<td>109±7</td>
<td>80±4*</td>
</tr>
<tr>
<td>CO, L min⁻¹</td>
<td>6.2±0.3</td>
<td>5.0±0.2*</td>
</tr>
<tr>
<td>CO/BSA, L min⁻¹ m⁻²</td>
<td>3.2±0.2</td>
<td>3.0±0.1</td>
</tr>
<tr>
<td>TPR, mm Hg L min⁻¹</td>
<td>15.8±0.8</td>
<td>18.2±0.8*</td>
</tr>
<tr>
<td>TPR/BSA, mm Hg L min⁻¹ m⁻²</td>
<td>31.4±1.6</td>
<td>31.1±1.6</td>
</tr>
<tr>
<td>MSNA, bursts min⁻¹</td>
<td>25±1</td>
<td>19±3*</td>
</tr>
<tr>
<td>MSNA, bursts 100 heartbeats⁻¹</td>
<td>44±2</td>
<td>32±5*</td>
</tr>
</tbody>
</table>
burst frequency, although relationships were somewhat weaker. These data are also included in text form below.

MAP did not correlate with MSNA in men or women when MSNA was expressed as burst frequency or burst incidence (Figure 1; P > 0.05). As expected, TPR was positively related with MSNA in men when described as burst frequency (Figure 2) and burst incidence (r = 0.58; P < 0.05). The correlation between MSNA and TPR was similar when TPR normalized for body surface area (burst frequency: r = 0.55; burst incidence: r = 0.59; P < 0.05 for both). In complete contrast, in women, TPR was inversely related to MSNA (r = −0.49; P < 0.05), and there was a trend toward a negative relationship when MSNA was measured as burst incidence (r = −0.40; P = 0.09). However, 1 female participant exhibited an unusually high MSNA and a low TPR (Figure 2, circled point), which appeared to drive the negative correlation. When this subject was removed from the analysis, there was no relationship between TPR and MSNA in women (burst frequency: r = −0.27; burst incidence: r = −0.17; P > 0.05). Moreover, the lack of relationship between MSNA and TPR in women persisted when TPR normalized for body surface area was used in the analysis (burst frequency: r = −0.23; burst incidence: r = −0.21; P > 0.05 for both).

Resting cardiac output in the young men was inversely related to MSNA recorded as both burst frequency (Figure 3) and burst incidence (r = −0.62; P < 0.05). The relationship between MSNA and cardiac output in men was similar when the cardiac output normalized for body surface area (ie, cardiac index) was used in the analysis (burst frequency: r = −0.67; burst incidence: r = −0.64; P < 0.01 for both). In the young women, however, cardiac output was positively correlated with MSNA (burst frequency: r = 0.52; burst incidence: r = 0.41; P = 0.1). However, when the female participant with a high resting MSNA was removed from the analysis (Figure 3, circled point), there was no correlation between MSNA and cardiac output (burst frequency: r = 0.23; burst incidence: r = 0.12; P > 0.05 for both). In addition, the lack of relationship between MSNA and cardiac output persisted in women when cardiac output was normalized for body surface area (ie, cardiac index; burst frequency: r = 0.22; burst incidence: r = 0.08; P > 0.05).

To explore the relationship between MSNA and cardiac output, we examined whether MSNA was related to stroke volume and/or heart rate. Stroke volume was inversely correlated with MSNA expressed as bursts frequency in men (Figure 4), whereas when MSNA was expressed as burst incidence only, a trend toward a positive relationship existed (r = −0.39). In women (n = 17), there was a trend toward a negative correlation between stroke volume and MSNA (burst frequency: r = 0.44; burst incidence: r = 0.45; P = 0.07), which disappeared when the same female participant was removed from the analysis (burst frequency: r = 0.27; burst incidence: r = 0.18; P > 0.05). Heart rate was not related to MSNA in the men (burst frequency: r = 0.14; burst incidence: r = −0.03; P > 0.05) or women (burst frequency: r = −0.02; burst incidence: r = −0.20; P > 0.05).
Discussion

The major new finding of the present study is that the interindividual positive relationship between TPR and MSNA and the inverse relationship between cardiac output and MSNA observed previously in men do not exist in young normotensive women. In striking contrast to previous findings in men, we found that women had no relationship between MSNA and TPR (Figure 2). This observation suggests that, among the factors that contribute to the overall level of TPR, the magnitude of sympathetic nerve activity has a greater role in young men compared with young women. Therefore, other key factors may have a greater contribution to the control of TPR in resting women and may explain why women have less autonomic support of blood pressure than men.

MSNA as an Index of Sympathetic Activity

Several lines of evidence support the idea that at-rest MSNA is a good indicator of the net vasoconstrictor tone directed toward the vasculature in young healthy men. In addition to our previous study, other work demonstrates that at-rest MSNA is well correlated to total arterial norepinephrine spillover, cardiac and renal norepinephrine spillover, and arterial plasma norepinephrine in normotensive healthy volunteers. However, all of these studies were conducted in men. Our present study suggests that MSNA may not be a good indicator of vasoconstrictor tone in the peripheral vasculature of resting normotensive young women. Furthermore, physiological stressors, such as orthostasis, and pathophysiology, such as obesity or hypertension, add additional complexity to the relationship between MSNA and vasoconstrictor tone, so it is less clear whether MSNA is closely related to cardiac, renal, or whole body norepinephrine spillover in those conditions.

Possible Mechanisms Contributing to the Dissociation of MSNA and TPR in Women

Several mechanisms may contribute to the lack of relationship between sympathetic nerve activity and TPR in women. First, estrogen has a direct vasodilator effect on the vasculature, which might compete with sympathetic vasoconstriction. Second, estrogen appears to increase the bioavailability of NO, which, again, might offset sympathetic vasoconstriction. Third, estrogen supplementation in rats increases vasodilating \( \beta_2 \)-adrenergic receptor–mediated responses to isoproterenol. Moreover, in women, vasoconstrictor responses to norepinephrine are enhanced by concurrent administration of \( \beta \)-blockers, which also eliminates the difference in \( \alpha \)-mediated vasoconstrictor responses between men and women. Therefore, \( \alpha \)-adrenergic vasoconstriction associated with a given sympathetic neural impulse might be offset by greater \( \beta_2 \)-adrenergic–mediated vasodilatation in women. Other possibilities that might contribute to the lack of relationship between MSNA and TPR in women include less

![Figure 3. Linear regression analysis of the relationship between cardiac output (nonnormalized CO) and MSNA in men and women. The relationship between CO and MSNA in women suggests that when MSNA is high CO is elevated, which is opposite to the relationship reported in men. However, once the circled female outlier was removed there was no correlation between MSNA and CO in women (burst frequency: \( r = 0.23; \) burst incidence: \( r = 0.12; P > 0.05 \)).](image1)

![Figure 4. Linear regression analysis of the relationship between stroke volume (SV) and MSNA in men and women. There was no correlation between SV and MSNA when the circled female participant was removed from the analysis (burst frequency: \( r = 0.27 \); burst incidence: \( r = 0.30; P > 0.05 \)).](image2)
norepinephrine and/or neuropeptide Y release per burst of sympathetic traffic, which might be modified by the effect of estrogen and progesterone on the sympathetic nerve terminals. Alternatively, higher circulating levels of endogenous nonadrenergic vasoconstrictors may alter the relationship between MSNA and TPR in women. However, there is inadequate information in the literature to comment definitively on these possibilities.

Consistent with the ideas mentioned above, loss of the vasodilatory effects of female sex hormones may also explain why postmenopausal women with high baseline MSNA have increased blood pressure. Although our present study was not designed to test the specific effects of estrogen on peripheral vascular tone, our data are consistent with these previous reports. They also raise important new questions about sex differences and fundamental integrative mechanisms regulating arterial pressure in humans.

Relationship Between MSNA and Cardiac Output
The lack of relationship between MSNA and arterial pressure has proven to be perplexing, because sympathetically mediated vasoconstriction would be expected to increase arterial pressure. In young men, this paradox can be explained in part by the balance that appears to exist between MSNA and cardiac output. That is, because MSNA contributes to TPR, and arterial pressure is a balance between cardiac output and TPR (ie, MAP = cardiac output x TPR), the inverse relationship between MSNA and cardiac output contributes to the normal blood pressure in young men with higher MSNA.

Our present observation of no relationship between MSNA and cardiac output in women suggests that the sexes rely on different mechanisms to maintain a normal arterial pressure regardless of widely varying MSNA. In the present study, we also noted that MSNA was inversely related to stroke volume in men, but again this relationship was not evident in the women (Figure 4). One possible mechanistic explanation for these observations is that augmented β2-adrenergic responsiveness because of sex hormone influences also extends to β2-adrenergic control of cardiac myocyte contractility. Another potential explanation is that differences in afterload for a given MSNA cause the differences in the relationship between stroke volume and MSNA among young men and women. In other words, men with a low MSNA have a relatively lower TPR and, therefore, a lower afterload, which reduces the opposition to the outflow of blood from the left ventricle; therefore, stroke volume is elevated in men with low MSNA. This does not appear to occur in young normotensive women, because TPR is not related to MSNA.

Other Observations
We noted a number of other differences between men and women. Mean MSNA, MAP, and cardiac output were lower in women than men, whereas TPR was higher in women than in men, which is consistent with previous studies. However, when we scaled cardiac output and TPR for body surface area, the sex differences in cardiac output and TPR were abolished. Therefore, differences in TPR between men and women are likely a result of differences in body size and its effect on cardiac output.

Conclusions
In summary, the results of the present study demonstrate that the interindividual relationships among TPR, cardiac output, and MSNA observed previously in young men do not exist in young women. Thus, in young women, sympathetic nerve activity does not determine TPR, and cardiac output is not balanced with MSNA to maintain normal arterial pressure. Consequently, it appears that arterial pressure regulation is fundamentally different between the sexes; this may be because of influences of female reproductive hormones on cardiovascular function. These differences in arterial pressure regulation may also explain sex differences in the prevalence of orthostatic intolerance in young women and why older women with high levels of MSNA have increased arterial pressure.

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Limitations
There are several important limitations to our study. First, we measured cardiac output noninvasively. However, we used a nonbreathing acetylene uptake technique that has a reported variability of ~7%, which is similar to other invasive techniques and noninvasive approaches to measuring cardiac output. Second, individual blood estrogens and progesterone levels were not measured in this study; thus, we can only speculate that estrogen-mediated effects on the vasculature are a major explanation for our findings, but this remains a major hypothesis going forward from this study and is certainly consistent with existing data.

Perspectives
For many years, the interindividual variability in sympathetic neural activity in humans was thought to limit our ability to understand the role of sympathetic neural mechanisms in arterial pressure regulation. Our recent work and that of others indicates that the opposite appears to be true. Exploring how interindividual variabilities in key hemodynamic variables interact as determinants of blood pressure provides key insights into integrative regulatory mechanisms. Moreover, our observations may have important clinical implications. First, the differences that we observed between men and women in the present study may provide insights into clinically relevant sex differences, ranging from greater orthostatic intolerance in young women to greater hypertension in young men. Second, as humans age, the relationship between MSNA and MAP becomes positive more so in women than in men. This raises the possibility that, as the estrogen-related effects that we mention are lost in postmenopausal women, those with high levels of MSNA will be at a greater risk for hypertension. In this context, future studies regarding the influence of aging on the balance among sympathetic nerve activity, central hemodynamics, and peripheral vasoconstrictor tone will likely further our understanding of the role played by the female sex hormones in blood pressure regulation.
conduct of the studies and analysis of the data. Finally, we thank the subjects for their participation.

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Disclosures
None.

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