

Adiposity Is Related to Gender Differences in Impaired Stress-Induced Pressure Natriuresis

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Abstract—The purpose of this study was to determine if there are gender differences in stress-induced pressure natriuresis and to examine the effects of adiposity on these differences. The subjects were 151 boys and 141 girls 15 to 18 years of age who underwent a 5-hour stress protocol (2-hour prestress, 1-hour stress, 2-hour poststress) after being brought into similar levels of sodium balance. The gender-by-condition interaction was significant for systolic and diastolic blood pressure ($P=0.001$ for both), and the effect of condition was significant for sodium excretion ($P=0.001$). Systolic blood pressure was higher for boys throughout the protocol ($P=0.001$ for each) and correlated with body mass index at each condition (range in $r=0.28$ to 0.35 ; $P<0.001$ for each). Hemodynamically, in boys body mass index was correlated with cardiac output during stress ($r=0.23$; $P=0.006$), which was correlated with systolic blood pressure ($r=0.21$; $P=0.01$). With respect to natriuresis, body mass index was inversely correlated with sodium excretion during stress ($r=-0.22$; $P=0.008$) and positively correlated with angiotensin II in a subsample of boys ($n=89$; $r=0.31$; $P=0.003$). The inverse correlation between angiotensin II and sodium excretion during stress approached significance ($r=-0.17$; $P<0.06$). Similar results were not observed for girls. In conclusion, gender differences in stress-induced pressure natriuresis appear to be related to the influence of adiposity on both blood pressure and natriuresis. (*Hypertension*. 2003;42:1082-1086.)

Key Words: gender ■ obesity ■ stress ■ sodium ■ blood pressure ■ natriuresis ■ hypertension, sodium-dependent

We hypothesized¹ that impaired stress-induced pressure natriuresis contributes to the development of essential hypertension (HTN) and blood pressure (BP)-related target organ damage by increasing the cardiovascular load experienced as a result of the stress. Our definition of impaired stress-induced pressure natriuresis is an increase in BP during an extended period of stress without an adequate compensatory increase in urinary sodium excretion ($U_{Na}V$). We provided indirect support for our hypothesis in a recent study¹ in which black compared with white youths had slower natriuresis across a series of stressors. This pattern was associated with cardiac remodeling. We provided additional support for our hypothesis in another recent study² that demonstrated that a significant percentage of black youths showed impaired stress-induced pressure natriuresis during an extended stress period, with BP remaining elevated as long as 2 hours after the cessation of the stressor. Subsequently, we reported³ that obese compared with lean youths had slower natriuresis during an extended stress period despite similar levels of BP.

The well-documented gender differences in cardiovascular disease and HTN have received increased attention in recent years (for recent reviews, see References 4 through 7)⁴⁻⁷. Furthermore, gender differences in cardiovascular responses

to acute stress have long been recognized.^{8,9} The first goal of this study was to determine if there are gender differences in changes in BP and $U_{Na}V$ during extended stress. A second goal was to determine if these differences are related to adiposity. A final goal was to examine the roles of angiotensin (Ang) II and insulin on the response patterns in a subgroup of boys and girls.

Methods

Study Population

The protocol was approved by the Human Assurance Committee of the Medical College of Georgia. Written informed parental consent and subject assent were obtained before testing. The subject characteristics are provided in Table 1. The range in body mass index (BMI) for boys was 15.78 to 39.84 kg/m² compared with 16.01 to 45.0 kg/m² for girls. Approximately 24% ($n=36$) of the boys and 46% ($n=65$) of the girls were either overweight or obese as defined by a BMI >25 kg/m².

Protocol

Our protocol has been described previously in detail.^{2,3} Briefly, the subjects were placed on a controlled sodium diet of 4000 ± 200 mg/d for 3 days before testing.¹⁰ On the fourth day, the subjects performed the stress protocol. The stress protocol included a pretest period of 2 hours, during which the subjects watched movies and/or listened to

Received July 2, 2003; first decision July 21, 2003; revision accepted September 9, 2003.

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Hypertension is available at <http://www.hypertensionaha.org>

DOI: 10.1161/01.HYP.0000100424.84889.F7

TABLE 1. Subject Characteristics and Casual Measurements by Gender and for the Group as a Whole

Variable	Boys (n=151)	Girls (n=141)	Total (n=292)
Age, y	16±1	16±3	16±1
Height, m*	1.7±1	1.6±1	1.7±1
Weight, kg	71±15	69±20	70±18
Body mass index, kg/m ² †	23±4	26±7	24±6
Casual systolic blood pressure, mm Hg*	109±8	105±10	108±10
Casual diastolic blood pressure, mm Hg	67±7	67±8	67±8
Baseline angiotensin II, pg/mL‡	13±7	16±12	15±10
Baseline insulin, U/mL‡	40±35	57±32	50±34

*Boys greater than girls, $P<0.001$; †girls greater than boys, $P<0.001$; ‡girls greater than boys, $P<0.05$.

the radio. This was followed by a 1-hour stress period, during which the subjects played a competitive video game task for a monetary reward (Snowboard, Sony Corp). Finally, there was a 2-hour posttest "recovery" period that was the same as the pretest period. Urine and blood samples were obtained hourly, and hemodynamic measurements were obtained at 15-minute intervals with the Dinamap monitor (Dinamap Compact Monitor) for BP and the Minnesota Impedance Cardiograph (MIC: Model 304B) for stroke volume (SV), cardiac output (CO), and total peripheral resistance (TPR). The subjects were required to drink 200 mL of water every hour to ensure they remained hydrated and provided adequate urine samples.

Measurement of Adiposity

Weight and height (in shorts and tee-shirt, without shoes) was measured with an electronic scale and stadiometer. These values were used to calculate BMI ($\text{BMI}=\text{wt}/\text{height}^2$), which was used as a measure of general adiposity.

Assays

Angiotensin II

Corresponding Ang II measurements were available for 89 boys and 81 girls. Ang II was evaluated by RIA. Plasma was extracted with HPLC-grade methanol on phenylsilylsilica extraction columns. The sample was reconstituted in Tris buffer. Ang II was measured by RIA with a kit (Buhlmann) distributed by ALPCO. The minimum detectable level of the assays was 2 pg/mL for Ang II. The intra-assay coefficient of variation was <10% for Ang II. The interassay coefficient of variation was 20.7%.

Insulin

Corresponding insulin concentrations were available for 31 boys and 41 girls. Insulin was measured in duplicate by a solid-phase, antibody-coated-tube RIA with kits purchased from Diagnostic Products Corp. The antibody cross-reacts 20% with proinsulin and exhibits nondetectable cross-reactivity with C-peptide and glucagon. Intra-assay coefficient of variation was <10% and interassay variation was 14.7%.

Analysis

The analyses were performed with SPSS 11.0. A repeated-measures ANOVA was performed to determine the effects of hour/condition (hour 1, hour 2, stress, hour 4, hour 5) and gender on systolic BP (SBP), diastolic BP (DBP), $U_{Na}V$, insulin, and Ang II throughout the 5-hour protocol. Significant differences in the models were determined by means of the Bonferroni test, which adjusts for multiple comparisons. Pearson correlation coefficients were used to examine the relationships between the variables for boys and girls separately.

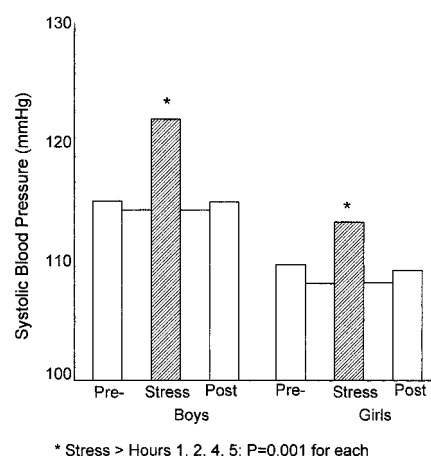


Figure 1. Changes in systolic blood pressure throughout 5-hour protocol for boys (n=151) and girls (n=141).

Results

Subject Characteristics

The subject characteristics and casual measurements are provided in Table 1. Boys compared with girls were taller and had higher casual SBP. The girls had a greater BMI and baseline levels of both insulin and Ang II. The sample for the girls was composed of a greater percentage of blacks (119/141=84%) than the boys (93/151=62%; $P=0.001$).

Gender Differences in Hemodynamic Functioning

The changes in BP across the test period for SBP are shown in Figure 1. The effect of condition ($P=0.001$) and the gender-by-condition interaction ($P=0.005$) were significant. The boys compared with the girls had higher SBP throughout the protocol ($P=0.001$ for each). Blood pressure during the stress period (hour 3) was significantly higher than any of the other hours for both boys and girls, with a significant increase from baseline hour 2 to stress and a significant decrease from stress to recovery hour 1, as shown in Figure 1 ($P=0.001$ for each). The boys had a greater stress-induced increase and poststress-related decrease in SBP ($P=0.001$ for both). Hemodynamically, SBP was correlated with CO during stress ($r=0.21$; $P=0.01$). Similar results were observed for DBP in boys, with a significant effect of condition ($P=0.0001$) and gender-by-condition interaction ($P=0.001$). Boys also had a greater stress-induced increase ($P=0.001$) and poststress decrease ($P=0.01$) in DBP.

The effect of condition ($P=0.001$) and the gender by condition interaction were also significant for heart rate ($P=0.002$). The girls had higher heart rates throughout the protocol ($P=0.001$ for each), with similar changes in heart rate from baseline hour 2 to stress for the boys and girls.

Influence of Adiposity

Body mass index was correlated with SBP throughout the protocol in boys, as shown in Table 2. Hemodynamically, BMI was correlated with CO during stress ($r=0.23$; $P=0.006$) in boys. In contrast, BMI was only correlated with SBP during the second hour of recovery in girls. Body mass

TABLE 2. Correlation Coefficients Between Body Mass Index and Systolic Blood Pressure Throughout the Protocol

Time	Boys	Girls
Hour 1	0.28*	0.10
Hour 2	0.28*	0.05
Hour 3 (stress)	0.30*	0.13
Hour 4	0.32*	0.15
Hour 5	0.35*	0.18†

* $P=0.001$; † $P=0.03$.

index was not correlated with DBP at any time point for either boys or girls.

Sodium Excretion

The data for $U_{Na}V$ are presented in Figure 2. The effect of condition was significant ($P=0.0001$), but the gender-by-condition interaction was not. Both boys and girls showed significant stress-related increases and poststress-related decreases in $U_{Na}V$.

Influence of Adiposity

Body mass index was inversely related to $U_{Na}V$ during the stress period in boys, as shown in Figure 3. The magnitude of the correlation became greater when only boys with a BMI >25 were considered ($r=0.29$; $P=0.04$). In contrast, the correlation between BMI and $U_{Na}V$ in girls was not significant.

Relationships With Percentile for Body Mass Index Adjusted for Age

The BMI adjusted for age and sex was determined by means of the SAS Program for the CDC Growth Charts. The correlations between percentile BMI and SBP were significant for hour 1 ($r=0.13$; $P=0.03$), hour 3 (stress: $r=0.14$; $P=0.04$), hour 4 ($r=0.16$; $P=0.007$), and hour 5 ($r=0.19$; $P=0.001$). The inverse correlation with stress $U_{Na}V$ approached significance ($r=-0.1$; $P=0.08$).

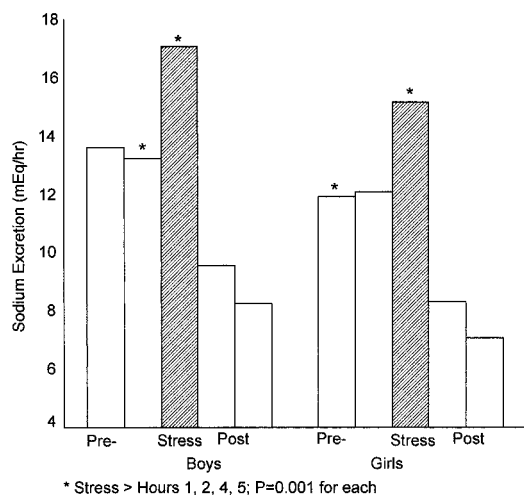


Figure 2. Changes in urinary sodium excretion throughout 5-hour protocol for boys ($n=151$) and girls ($n=141$).

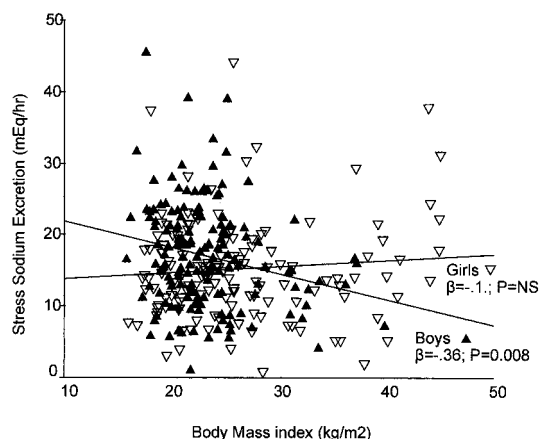


Figure 3. Relationship between body mass index and sodium excretion during stress for boys ($n=151$) and girls ($n=141$).

Adiposity and Hormonal Regulation

The main effect of condition and the gender by condition interaction were not significant for Ang II. The correlation between BMI and Ang II was significant in boys ($r=0.31$; $P=0.003$) but not girls. The inverse correlation between Ang II and sodium excretion during stress approached significance ($r=-0.17$; $P=0.06$). The main effect of condition was significant for insulin ($P=0.001$), which showed a progressive decline throughout the study in both boys and girls. Body mass index was not associated with insulin throughout the study in either sex. In addition, insulin was not associated with $U_{Na}V$ throughout the study in either sex.

Discussion

The major finding of this study is that the boys had a blunted natriuretic response to the increase in BP produced by a period of extended stress. Furthermore, this response pattern was related to adiposity. In contrast, girls showed the expected natriuretic response to the increase in BP during stress, which did not appear to be influenced by adiposity.

The observation of sexual dimorphism in stress-induced pressure natriuresis is consistent with a large body of literature that demonstrated higher BP levels and greater cardiovascular risk in male subjects.⁴⁻⁷ These results are also consistent with a growing literature indicating that male subjects show greater salt sensitivity than female subjects, as recently reviewed by Ely et al,¹¹ Dubey et al,⁵ and Reckelhoff.⁴ The findings of a study by Uehara et al¹² are particularly relevant to the current study because they examined the effect of adiposity on gender differences in BP. The study was based on data obtained during medical examinations in 7279 Japanese university students. Multivariate analyses indicated that BMI was an independent predictor of SBP in male students with a hypertensive father. A similar relationship was not observed in the female students.

Our findings that BMI is related to stress-induced pressure natriuresis is consistent with our previous study³ that examined the association between percent body fat as determined by dual-energy x-ray absorptiometry and stress-induced pressure natriuresis. In this study, we observed that obese ($>25\%$ body fat) compared with lean (12% to 20% body fat) subjects

had similar stress-induced increases in BP but lower $U_{Na}V$. These results are also consistent with previous studies in humans and several animal models of obesity that demonstrated that obesity-related HTN is characterized by a shift in the pressure-natriuresis curve, that is, impaired pressure-natriuresis.^{13–15} Studies demonstrating “salt sensitivity” in obesity provide further evidence of impaired pressure-natriuresis in obesity. One study is particularly relevant to the current study because it was performed in adolescents. Rocchini¹⁶ demonstrated that the pressure-natriuresis slope was positive, indicating salt sensitivity, and shifted to a higher level of BP for obese compared with nonobese subjects on a high but not a low sodium diet. However, a minimal weight loss normalized the pressure-natriuresis relation.

Hemodynamically the changes and level of SBP in the boys with the greater adiposity was volume-mediated rather than resistance-mediated. These results are consistent with the well-recognized hemodynamic differences between BP regulation in lean compared with obese individuals.^{17–19}

We observed the hypothesized association between Ang II and pressure natriuresis in boys but not girls. These findings are consistent with the well-established relationship between obesity and the renin-angiotensin system.^{13,14,20} These results are also consistent with a study by Schneider et al²¹ on stress-induced pressure natriuresis. This was a double-blind, randomized, crossover study that examined the mechanisms related to impaired sodium regulation in 66 normotensive subjects (half of them with a family history of HTN) and 36 subjects with mild HTN. The subjects were studied at rest and during a 30-minute mental stress test, while treated with either a placebo or an ACE inhibitor. They observed a blunted increase in $U_{Na}V$ during the stress period in the patients with HTN as compared with normotensive subjects. The normotensive patients with a positive family history also showed a blunted increase as compared with those without a family history, although the difference was not significant. ACE inhibition normalized the response pattern in the patients with HTN.

We did not observe the hypothesized association between BMI and insulin levels throughout the protocol in either the boys or the girls. In addition, we did not observe the hypothesized association between insulin and $U_{Na}V$ during stress. Insulin showed a progressive decline throughout the protocol, whereas $U_{Na}V$ increased during stress and decreased after stress in both boys and girls. The correlations between insulin and $U_{Na}V$ were not significant at any time during the protocol. These results are in contrast to the well-described antinatriuretic actions of insulin under nonstress conditions.^{22–24}

The factors responsible for the gender differences in stress-induced pressure natriuresis and the gender difference in the effect of adiposity on stress-induced pressure natriuresis remain to be identified. Gender differences in cardiovascular responses to acute stress have long been recognized.^{8,9} These differences have been attributed to a number of potential factors ranging from differences in effort and enthusiasm to the protective effects of female sex hormones. We observed similar levels of self-reported effort and enthu-

siasm during the stress period. We did not measure sex hormones, so this remains a viable hypothesis. A third possibility is that the range of BMI for the different groups was not large enough to observe significant associations.

Perspectives

Male subjects have less body fat than female subjects and yet have greater obesity-related cardiovascular disease than premenopausal female subjects. The reason for this apparent paradox is not clear. The results of the present study provide evidence to suggest that stress may be a contributing factor through its effect on pressure natriuresis. Specifically, our data suggest that adiposity plays a greater role in the stress-induced pressure natriuresis response in boys than girls. This conclusion is tempered by the fact that the sample of boys in this study did not include a large number of obese individuals and therefore it is difficult to generalize the results to the population. As such, these findings need to be replicated in a cohort of subjects composed of a similar number of boys and girls with both low and high BMIs. The physiological reason for a greater effect of adiposity on stress-induced pressure natriuresis in male subjects is unclear and needs to be explored in future studies. One possibility is that the effect is due to the influence of sex hormones. Specifically, estradiol in the girls may blunt the sympathetically mediated increase in BP and/or increase vasodilation by stimulating the release of nitric oxide. Alternatively, androgens in boys may augment the BP response. A third possibility is that factors derived from the increased adipose tissue in girls such as leptin and angiotensinogen serve to promote natriuresis. This alternative is supported by the results of a very interesting recent study²⁵ in which male, female, and testosterone-treated female rats were fed a normal diet supplemented with a “cafeteria” diet composed of human foods including cheese, ham, sausage, potato chips, and candies. Fat mass increased in each group and was accompanied by the development of HTN in both the males and the testosterone-treated females and was associated with downregulation of the renal leptin receptor.

Acknowledgments

This study was supported by grants HL-59954 and HL-64225 from the National Institutes of Health.

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