Influence of plasma osmolality on baroreflex control of sympathetic activity

Megan M. Wenner,1 William C. Rose,2 Erin P. Delaney,1 Michael E. Stillabower,1,3 and William B. Farquhar1,2,3

1Department of Health, Nutrition, and Exercise Sciences and 2College of Health Sciences, University of Delaware; and 3Christiana Care Health Systems, Cardiovascular Research, Newark, Delaware

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Wenner MM, Rose WC, Delaney EP, Stillabower ME, Farquhar WB. Influence of plasma osmolality on baroreflex control of sympathetic activity. Am J Physiol Heart Circ Physiol 293: H2313–H2319, 2007. First published July 20, 2007; doi:10.1152/ajpheart.01383.2006.—The purpose of this study was to determine if plasma osmolality alters baroreflex control of sympathetic activity when controlling for a change in intravascular volume: we hypothesized that baroreflex control of sympathetic activity would be greater during a hyperosmotic stimulus compared with an isosmotic stimulus when intravascular volume expansion was matched. Seven healthy subjects (25 ± 2 yr) completed two intravenous infusions: a hypertonic saline infusion (HSI; 3% NaCl) and, on a separate occasion, an isotonic saline infusion (ISO; 0.9% NaCl), both at a rate of 0.15 ml kg⁻¹ min⁻¹. To isolate the effect of osmolality, comparisons between HSI and ISO conditions were retrospectively matched based on hematocrit; therefore, baroreflex control of sympathetic outflow was determined at 20 min of a HSI and 40 min of an ISO. Muscle sympathetic outflow (MSNA) was directly measured using the technique of peroneal microneurography; osmolality and blood pressure (Finometer) were assessed. The baroreflex control of sympathetic outflow was estimated by calculating the slope of the relationship between MSNA and diastolic blood pressure during controlled breathing. Plasma osmolality was greater during the HSI compared with the ISO (HSI: 292 ± 0.9 mosmol/kg and ISO: 289 ± 0.8 mosmol/kg, P < 0.05). Hematocrits were matched (HSI: 39.1 ± 1% and ISO: 39.1 ± 1%, P > 0.40); thus, we were successful in isolating osmolality. The baroreflex control of sympathetic outflow was greater during the HSI compared with the ISO (HSI: −8.3 ± 1.2 arbitrary units·beat⁻¹·mmHg⁻¹ vs. ISO: −4.0 ± 0.8 arbitrary units·beat⁻¹·mmHg⁻¹, P = 0.01). In conclusion, when controlling for intravascular volume, increased plasma osmolality enhances baroreflex control of sympathetic activity in humans.

baroreceptor; muscle sympathetic nerve activity; intravascular volume

recently, data from both experimental animals and humans have suggested that sympathetic outflow is influenced by plasma osmolality (2, 4, 12, 29). For example, in a water-deprived rat model (where plasma osmolality and lumbar sympathetic nerve activity are initially high), the administration of a 5% dextrose in water solution caused a parallel decline in plasma osmolality and lumbar sympathetic outflow (29), supporting a relationship between plasma osmolality and sympathetic outflow. This osmotic-sympathetic interaction has also been demonstrated in humans (12). An infusion of hypertonic saline (HSI), resulting in an increase in both plasma osmolality and intravascular volume, was associated with an acute increase in basal muscle sympathetic nerve activity (MSNA); however, the increase in sympathetic activity did not parallel the increase in osmolality, as MSNA declined despite further elevations in plasma osmolality and intravascular volume expansion (12). The baroreflex control of sympathetic outflow has been examined during a HSI in humans (4). Charkoudian et al. (4) reported that relatively small increases in osmolality increase baroreflex control of sympathetic outflow; however, despite further increases in osmolality (with a faster infusion rate), there were not additional increases in sympathetic baroreflex sensitivity. The reason for the transient excitatory effects during a progressive HSI is not clear.

One possible explanation for the waning effects of HSI to increase basal MSNA and arterial baroreflex function may be that the concurrent volume expansion counteracts the effects of osmolality per se. Indeed, changes in intravascular volume can have direct effects on the baroreflex control of sympathetic outflow: the decline in intravascular volume during prolonged bed rest is associated with increases in sympathetic baroreflex sensitivity (22). Similarly, the decline in central blood volume during head-up tilt is associated with increases in sympathetic baroreflex sensitivity (13). In contrast, increases in intravascular volume are associated with decreases in baroreflex control of sympathetic activity. For example, increases in central venous pressure, either with saline infusion or acute head-down tilt, are associated with decreases in sympathetic baroreflex sensitivity (7). These studies (4, 5, 7, 13, 22) support an inverse relationship between volume status and sympathetic baroreflex sensitivity.

The above-mentioned volume-induced changes in sympathetic baroreflex function make it difficult to isolate the effects of osmolality, since the increases in sympathetic outflow induced by HSI (4, 12) may be partially opposed by the simultaneous changes in volume. For example, elevations in plasma osmolality are associated with exercise-associated intravascular volume depletion; an isotonic saline infusion (ISO; i.e., intravascular volume expansion) after exercise elicited a decrease in muscle sympathetic outflow and tended to decrease sympathetic baroreflex sensitivity (5).

The purpose of this study was to isolate the effects of increases in plasma osmolality on arterial baroreflex control of sympathetic activity. To address this question, we compared the effect of a hyperosmotic stimulus (HSI) with that of an isosmotic stimulus (ISO) during conditions where intravascular volume expansion was matched. Using this approach, we experimentally isolated plasma osmolality; this was done in a retrospective fashion. We hypothesized that baroreflex control of sympathetic outflow would be greater when osmolality was elevated.

METHODS

This is a retrospective analysis from previously published data (12). Seven young, normotensive subjects (6 men and 1 woman, age: 25 ±

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2 yr, body mass index: 24 ± 0.7 kg/m²) completed both a HSI (3% NaCl) and, on a separate occasion, an ISO (0.9% NaCl). Each subject completed both infusions. Before participation, all subjects provided verbal and written informed consent. The study protocol was approved by the Institutional Review Board of the University of Delaware.

**Screening session.** Prior to the first infusion, each subject was screened to ensure they were free from disease. After a 12-h fast (food, alcohol, caffeine, and exercise), blood was drawn to analyze cholesterol (triglycerides, HDL-cholesterol, LDL-cholesterol, and total cholesterol), electrolytes (sodium, potassium, and chloride), and enzymes for liver (aspartate transaminase and alanine transaminase) and kidney (blood urea nitrogen and creatinine) function. All blood results from the screening session were within clinically acceptable limits. Height and weight were measured (Heathymeter scale) and used to calculate the body mass index for all subjects. Each subject completed a physical activity readiness questionnaire and a medical history form. Blood pressure (BP) and heart rate (HR) were measured with a mercury sphygmomanometer and 12-lead ECG (Schiller AT-10, Electra-Med, Flint, MI), respectively, at rest and during a submaximal exercise test using an electronically braked cycle ergometer (Corival V2 Ergometer, Lode B.V. Medical Technology). Resting BPs confirmed that all of the participating subjects were normotensive (8). The exercise test began with a 2-min warm-up; the initial resistance was set at 75 W and increased 25 W every 2 min until 85% of the age-predicted maximal HR was reached. Resting and exercise ECGs were normal. All subjects were free from using tobacco and taking any medications, including oral contraceptives for the female subject.

**Experimental protocol.** On the day before the infusion, subjects were instructed to avoid salty foods and to consume ~1,800 ml of water throughout the day. They were also instructed to consume another 600 ml of water on the morning of the infusion prior to coming into the laboratory and had fasted from food, alcohol, caffeine, and exercise for at least 12 h prior to the infusion. The female subject was tested during the early follicular phase of the menstrual cycle. Subjects reported to the laboratory at ~7:00 AM. Upon arrival, subjects were asked to completely empty their bladder. A urine sample was taken to analyze urine specific gravity and, for the female subject, to complete a pregnancy test, if appropriate. Subjects were instrumented with a single-lead ECG and standard upper arm automated BP cuff (Dinamap Dash 2000, GE Medical Systems, Milwaukee, WI). Elastic bands were placed around the chest and abdomen to measure respiration (Inductotrace System, Ambulatory Monitoring, Ardsley, NY). Beat-by-beat BP was assessed using the Finometer (Finapres Medical Systems). This technique is highly correlated with direct intra-arterial measurements (27) and is a reliable measure of arterial pressure during various autonomic stressors (20, 21). A 20-gauge intravenous catheter was placed into the vein of each arm in the antecubital fossa. The catheter in the left arm was used to infuse either 3% or 0.9% saline (Lifecare 5000 infusion pump, Abbott Laboratories, Chicago, IL), while the catheter in the right arm was used for blood sampling during the infusion. The administration of saline was determined at random, and subjects were blinded as to which percentage of saline was being infused during either visit. Saline was infused at a rate of 0.15 ml·kg⁻¹·min⁻¹ for a total of 60 min; infusions were separated by approximately 1 min. A controlled HSI has been demonstrated as a safe and effective way to acutely increase plasma osmolality (4, 12, 30).

MSNA was measured directly from the peroneal nerve via the technique of microneurography as previously described (10, 11, 31, 32, 35). A recording electrode (tungsten microelectrode) was inserted into the peroneal nerve behind the fibular head; a reference electrode was inserted 2–3 cm away on the lower leg. MSNA was measured in the same leg in each subject during the two sessions. The MSNA raw signal was amplified (70,000-fold), bandpass filtered (700–2,000 Hz), rectified (full), and integrated (time constant: 0.1 s) using a nerve traffic analyzer (model 662-4, Nerve Traffic Analysis System, University of Iowa Bioengineering, Iowa City, IA). Per the Eckberg and Sleight textbook (10), the following criteria were used to determine if the nerve activity was MSNA (i.e., not skin): 1) electrical stimulation with the microelectrode caused a visible muscle twitch, 2) light stroking of the skin did not elicit bursts, 3) passive stretch resulted in muscle afferent bursts, and 4) an increase in burst activity during straining (Valsalva). Because of the time involved in obtaining a suitable nerve recording, subjects were supine ~45 min before baseline measurements began.

HR, BP, and sympathetic outflow were measured and analyzed at baseline for 5 min prior to the commencement of the infusion. To minimize hemodynamic changes, these variables were recorded and analyzed during 5 min of paced breathing at 0.25 Hz (i.e., 15 breaths/min); subjects followed the breathing rate (which was visually confirmed) by listening to a prerecorded CD. Whole blood was collected (venous blood sample) in the appropriate vacutainer and spun in a centrifuge (Allegra X-22R, Beckman Coulter, Fullerton, CA) at 2,500 rpm for 15 min. Serum sodium, potassium, and chloride (EasyElectrolyte Analyzer, Medica, Bedford, MA) and plasma osmolality (3D3 Osmometer, Advanced Instruments, Norwood, MA) were analyzed in triplicate in our laboratory after quality controls were run. In addition, precalibrated capillary tubes were filled with whole blood for the analysis of hematocrit. These tubes were spun in triplicate on a Readacrit Centrifuge (Clay Adams Brand, Becton Dickinson, Parsippany, NJ). Whole blood was also transferred into collecting slides for the analysis of hemoglobin (Hemocue Hb 201+ analyzer, Hemocue, Lake Forest, CA).

**Data analysis.** HR, respiration, beat-by-beat BP, and MSNA were collected at 500 Hz using WINDAQ software (DATAQ Instruments, Akron, OH). During the 5 min of paced breathing throughout the infusion, signal processing software (CODAS, DATAQ Instruments) was used to peak detect the ECG, and the peak and valley detected the BP signals. Data (for HR, BP, and MSNA) were averaged within each 5-min paced breathing time point during the infusion (baseline ISO, baseline HSI, 20-min HSI, and 40-min ISO). Custom MatLab software (MatLab, The Math Works, Natick, MA) was used to analyze MSNA (16). The peak voltage signal (chosen by a representative large burst) was calibrated at 1,000 arbitrary units (AU), whereas the signal between bursts was calibrated at zero AU. The latency (time delay) from the R wave (of the QRS complex) to the sympathetic burst was measured in WINDAQ and entered into the MatLab program to ensure sympathetic bursts were in time with the cardiac cycle (i.e., bursts occurred during diastole) (31, 34). The average latency duration was 1.3 ± 0.02 s.

We examined two indexes of baroreflex control of sympathetic outflow: 1) the slope of the relationship between MSNA (expressed as AU per beat) and diastolic blood pressure (DBP); and 2) the slope of the relationship between MSNA (expressed as burst incidence) and DBP (6, 23, 24, 31), both during controlled breathing. A previous microneurographic study (31) demonstrated a strong correlation between MSNA and DBP and a weak correlation between MSNA and systolic blood pressure (SBP); since MSNA is modulated by DBP, this was used in the regression analysis to determine sympathetic baroreflex control (31). Data from the nerve recordings were binned over a 2-mmHg DBP range using the custom MatLab software mentioned, and the MSNA within a bin was presented. The MSNA was sorted into bins according to the DBP associated with each nerve activity burst. Mean MSNA (AU/beat) was computed for each pressure bin, and the mean DBP for all the beats in each bin was also computed. The beats in a bin spanning from 60 to 62 mmHg, for instance, would have a mean pressure somewhere between 60 and 62 mmHg. The linear regression of MSNA versus DBP was performed using mean nerve activities and mean DBPs from all the bins for each experimental run. Binning the data reduces the statistical impact of the inherent beat-by-beat variability in nerve activity from nonbaroreflex influences (4, 9, 13, 15, 31). When calculating the baroreflex control of sympathetic activity, only slopes with an r value of >0.50 were
Table 1. Blood and hemodynamic data

<table>
<thead>
<tr>
<th></th>
<th>Baseline During ISO</th>
<th>Baseline During HSI</th>
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<tbody>
<tr>
<td>Sodium, mmol/l</td>
<td>137.3 ± 1.2</td>
<td>136.9 ± 0.5</td>
</tr>
<tr>
<td>Potassium, mmol/l</td>
<td>3.91 ± 0.09</td>
<td>3.78 ± 0.07</td>
</tr>
<tr>
<td>Chloride, mmol/l</td>
<td>103.3 ± 1.7</td>
<td>103.9 ± 0.9</td>
</tr>
<tr>
<td>Hemoglobin, g/dl</td>
<td>15.2 ± 0.66</td>
<td>14.6 ± 0.5*</td>
</tr>
<tr>
<td>Heart rate, beats/min</td>
<td>63 ± 3</td>
<td>63 ± 3</td>
</tr>
<tr>
<td>Mean arterial pressure, mmHg</td>
<td>81 ± 2</td>
<td>86 ± 2*</td>
</tr>
<tr>
<td>Systolic blood pressure, mmHg</td>
<td>129 ± 4</td>
<td>138 ± 4*</td>
</tr>
<tr>
<td>Diastolic blood pressure, mmHg</td>
<td>65 ± 2</td>
<td>68 ± 2</td>
</tr>
<tr>
<td>Pulse pressure, mmHg</td>
<td>64 ± 3</td>
<td>70 ± 4*</td>
</tr>
</tbody>
</table>

Values are means ± SE. ISO, isotonic saline infusion; HSI, hypertonic saline infusion. ANOVA time: P < 0.05 for all variables except diastolic blood pressure, heart rate, and sodium. *P < 0.05, post hoc comparisons vs. the respective baseline.  

Accepted into the analysis (3, 28). The correlation coefficients from the linear regressions ranged from 0.70 to 0.98. DBPs used for analyzing arterial sympathetic baroreflex control spanned 14 ± 1.6 mmHg.

To experimentally isolate the effect of osmolality, we retrospectively matched a group of subjects based on hematocrit; therefore, all data were analyzed at 20 min of the HSI and 40-min of the ISO. Changes in volume status can be estimated from changes in hematocrit during various stressors (such as exercise) that do not last longer than 2 h and when plasma osmolality changes are 13 mosmol/kg H2O (14). Initially, there were eight subjects in the analysis; however, one subject was not included. During one of the infusion periods, there was a poor relationship between MSNA and DBP, and the r values were not acceptable based on the criteria previously stated. Therefore, we were not able to obtain a reliable index of baroreflex control of MSNA, and the subject was not included in the analysis.

Statistics. All data are reported as means ± SE. To assess arterial baroreflex control of sympathetic activity, a linear regression was performed between MSNA (AU/beat) and DBP (Sigma Plot 9.0) during paced breathing; the slope of the line determined by the regression analysis was used as an index of baroreflex control of MSNA. In addition, a linear regression was performed between MSNA (bursts/100 heart beats) and mean DBPs of each bin corresponding with the appropriate cardiac cycle (6, 24, 31). Two-way repeated-measures ANOVA (SPSS 14.0) with Bonferroni post hoc tests (when P < 0.05 by ANOVA) were used to determine statistical significance (baseline ISO, baseline HSI, 40-min ISO, and 20-min HSI). Statistical significance was set at a level of P < 0.05.

RESULTS

Table 1 lists blood work results and hemodynamic data (from the Dinamap) at baseline and under ISO and HSI conditions. Hemoglobin declined significantly during both conditions (baseline vs. HSI and baseline vs. ISO, P < 0.05 for both) and was similar between HSI and ISO time points. Chloride was greater during HSI compared with baseline, with no differences during ISO. During HSI, potassium was lower compared with its respective baseline, whereas there was no change in potassium during ISO. There tended to be a main effect of time (P = 0.07 by ANOVA) for serum sodium. SBP and pulse pressure increased significantly during both HSI and ISO, whereas no changes occurred in DBP or HR during either condition. There was a significantly greater increase in mean arterial pressure during the ISO condition.

Hematocrit declined during both ISO and HSI conditions (ISO: 40.8 ± 1.2% to 39.1 ± 1% and HSI: 41.6 ± 1.3% to 39.1 ± 1%, P < 0.01 vs. the respective baseline; Fig. 1, A and B) and were matched between time points (39.1 ± 1% vs. 39.1 ± 1%, P > 0.40; Fig. 1, C). Plasma osmolality (Fig. 2, A and B) did not change during ISO (289 ± 1.1 to 289 ± 0.8 mosmol/kg H2O, P = 0.20) but significantly increased during HSI (288 ± 0.8 to 292 ± 0.9 mosmol/kg H2O, P < 0.01). Plasma osmolality was significantly greater during HSI versus ISO (292 ± 0.9 vs. 289 ± 0.8 mosmol/kg H2O, P < 0.05; Fig. 2, C). Hematocrit was matched between HSI and ISO, and osmolality was greater during HSI compared with ISO; thus, we were successful in experimentally isolating osmolality.

Figure 3 shows the linear relationship between MSNA and DBP (i.e., baroreflex control of sympathetic outflow) from one subject during both conditions (A, ISO; and B, HSI). The slope of the linear regression was less steep during ISO, whereas the slope became steeper during HSI. Individual and mean data regarding baroreflex control of sympathetic outflow are shown in Fig. 4. During ISO, baroreflex control of sympathetic outflow significantly decreases (−6.34 ± 1 vs. −4.0 ± 0.8 AU·beat−1·mmHg−1, P = 0.01). In contrast, sympathetic baroreflex control increased (−6.35 ± 1 vs. −8.3 ± 1.2 AU·beat−1·mmHg−1, P = 0.03) during HSI (Fig. 4, A and B). Thus, when the intravascular stimulus was matched and plasma osmolality was elevated, the baroreflex control of sympathetic outflow was greater (HSI vs. ISO time points: −8.3 ± 1.2 vs. −4.0 ± 0.8 AU·beat−1·mmHg−1, P = 0.01; Fig. 4, C). Basal sympathetic activity did not change (ANOVA time: P = 0.30) from baseline during ISO (14.5 ± 3 to 13.4 ± 3 bursts/min) or HSI (15.3 ± 3 to 19.4 ± 2 bursts/min), although there was a trend toward a difference (ANOVA interaction: P = 0.07). Results from the burst incidence analysis are shown in Fig. 5. There was a trend toward a significant interaction (P = 0.06 by ANOVA) between the slope of the burst incidence-DBP relationship during the ISO condition (baseline vs. 40-min ISO: −3.4 ± 0.6 vs. −1.7 ± 0.3 bursts·100 heart beats·1·mmHg−1) and HSI condition (baseline vs. 20-min HSI: −2.9 ± 0.8 vs. −3.12 ± 0.3 bursts·100 heart beats·1·mmHg−1).

DISCUSSION

The major finding of the present study is that osmolality, when controlling for intravascular volume, influences arterial baroreflex control of sympathetic activity. Since volume and sympathetic baroreflex function are inversely related (4, 5, 7, 13, 22), it has been difficult to isolate the effect of osmolality on sympathetic baroreflex function. Therefore, our results, along with
recently published data (4), reinforce the idea of osmolality per se having an effect on sympathetic baroreflex control.

An osmotic-sympathetic interaction has been previously demonstrated in both animals and humans (12, 29). Osmoreceptors in the circumventricular organs of the brain may have a neuronal link with efferent sympathetic pathways (33). When examining this osmotic-sympathetic interaction, Scrogin et al. (29) demonstrated that a 1% decline in osmolality via dextrose infusion resulted in a 5% decrease in sympathetic outflow. In addition, small increases in osmolality resulted in an increase in baroreflex control of sympathetic activity (4).

As noted by Charkoudian et al. (4), there appears to be an inverse relationship between volume and sympathetic baroreflex sensitivity (4, 7, 22, 25): increases in central venous pressure either through ISO or head-down tilt decreased sympathetic baroreflex sensitivity (7). In contrast, hypovolemia (where intravascular volume is reduced) was associated with greater sympathetic outflow and baroreflex sensitivity at rest and during lower body negative pressure in humans (25). In addition, augmented baroreflex control of sympathetic activity was demonstrated after prolonged head-down bed rest (which is also associated with a decline in intravascular volume) (22).

Recently, Fu et al. (13) demonstrated an acute increase in baroreflex control of sympathetic activity during head-up tilt, again supporting an inverse relationship between sympathetic baroreflex sensitivity and volume. In our previous examination of osmotic-sympathetic interactions, sympathetic activity was attenuated (after an acute increase in burst frequency) despite...
a step-wise increase in plasma osmolality (12); this may have been from the volume load associated with the infusion. Thus, due to the confounding effects of volume, controlling the intravascular volume stimulus is of importance when examining the role of osmolality in baroreflex control of sympathetic activity. While the focus of the present analysis was on the effect of osmolality on baroreflex control of sympathetic activity during conditions of similar intravascular volume expansion, it is interesting to note that a failure to control for volume (40-min ISO vs. 40-min HSI) resulted in no difference in sympathetic baroreflex control ($P = 0.42$; data not shown). Had we only performed the HSI and not considered the potential confounding effects of volume, we might have concluded that elevations in plasma osmolality have a modest effect on sympathetic baroreflex control (Fig. 4, B) or we may have concluded, based on the burst incidence plot, that there was no effect (Fig. 5, B). Only by comparing the effect of a hyperosmotic stimulus to an isoosmotic stimulus during similar intravascular volume expansion (Fig. 4, C) do we see the strong effect of osmolality per se on the baroreflex control of sympathetic outflow.

Our analysis of baroreflex control of sympathetic outflow was performed during controlled breathing, where spontaneous BP changes occur. Although a high correlation between DBP

![Fig. 3. Linear relationship between muscle sympathetic nerve activity (MSNA; in arbitrary units (AU)/beat) and diastolic blood pressure (DBP; in mmHg) from 1 subject during the isotonic condition (A; ISO baseline: slope = -6.34, $r = 0.89$; ISO: slope = -4.5, $r = 0.83$) and during the hypertonic condition (B; HSI baseline, slope = -4.3, $r = 0.78$; HSI: slope = -8.8, $r = 0.83$).](image)

**Fig. 3.** Linear relationship between muscle sympathetic nerve activity (MSNA; in arbitrary units (AU)/beat) and diastolic blood pressure (DBP; in mmHg) from 1 subject during the isotonic condition (A; ISO baseline: slope = -6.34, $r = 0.89$; ISO: slope = -4.5, $r = 0.83$) and during the hypertonic condition (B; HSI baseline, slope = -4.3, $r = 0.78$; HSI: slope = -8.8, $r = 0.83$).

![Fig. 4. Baroreflex control of sympathetic outflow within isotonic (A) and hypertonic (B) conditions, displaying both individual and mean responses (ANOVA interaction: $P = 0.002$). During the isotonic condition, the slope of the relationship in 6 of 7 subjects decreased; there was a significant decline in sympathetic baroreflex control ($*P < 0.05$ vs. ISO baseline). In contrast, baroreflex control of MSNA significantly increased within the hypertonic trial ($*P < 0.05$ vs. HSI baseline); the slope of the relationship in 6 of 7 subjects either increased or did not change. The mean correlation coefficient from the individual linear regressions was $r = 0.89$. During conditions of similar intravascular volume expansion (InV) but with elevated osmolality (Osmo; C), the baroreflex control of sympathetic outflow was greater (HSI vs. ISO time points: $*P = 0.01$).](image)
and MSNA has been demonstrated during spontaneous BP fluctuations (31), larger changes in DBP are apparent when using the modified Oxford technique (9), thus allowing for the analysis of the entire sigmoid stimulus-response curve. Although our range of DBPs was somewhat narrow (14 mmHg), this approach is consistent with others in estimating sympathetic baroreflex control around the operating point and has been previously demonstrated during resting conditions and under various autonomic stressors (13, 19, 23, 24, 31).

**Limitations.** There were several limitations in the study. First, we did not assess arginine vasopressin (AVP). Due to the osmotic and volume stimuli from the infusions, fluid regulatory hormones such as AVP and plasma renin activity are important to quantify. It has been well established that AVP increases as a result of increased osmolality in humans (4, 30). The osmotic-induced release in AVP would be associated with sympathoinhibition and impaired reflex function (17). However, in rats, the changes in lumbar sympathetic nerve activity, which paralleled changes in osmolality, occurred independent of changes in vasopressin (29). Importantly, baroreflex control of sympathetic outflow was not altered when vasopressin was elevated in humans (9). Second, hematocrit is an imperfect index to estimate changes in intravascular volume. Based on Starling forces, fluid will move into the intravascular space when plasma osmolality is elevated; it is difficult to isolate the effects of osmolality without also influencing intravascular volume. Because of the composition of the saline infused, the hypertonic fluid could potentially shrink the red blood cells, thus overestimating the change in intravascular volume based on hematocrit. While a more appropriate index of changes in the intravascular volume may be to use plasma protein concentration (such as albumin), future studies may consider using this index of intravascular volume when isolating osmolality. Importantly, with the present analysis, hematocrit and hemoglobin values were both matched between hypertonic and isotonic conditions.

**Conclusions.** Our present findings support the role of plasma osmolality influencing baroreflex control of sympathetic outflow. This osmotic-sympathetic interaction may be an important neural mechanism contributing to cardiovascular control.

**Perspectives**

The influence of plasma osmolality on baroreflex control of sympathetic activity may be an important neural mechanism in cardiovascular control. The clinical relevance of this relationship is uncertain. During dehydration conditions, where there is a decline in intravascular volume and an increase in plasma osmolality, an increase in resting sympathetic outflow and an increase in baroreflex control of sympathetic outflow may serve as a protective mechanism against hypotension (4). This explanation makes teleological sense.

During dietary salt loading conditions, the relevance is also uncertain. Brooks et al. (1) has proposed that small increases in plasma sodium/osmolality trigger (through a central brain stem mechanism) sympathoexcitation. This sympathoexcitation may elevate BP and therefore be one of the mechanisms contributing to “salt-sensitive hypertension.” Ono et al. (26) reported that dietary salt loading enhanced arterial baroreceptor control of renal sympathetic nerve activity in normotensive Wistar-Kyoto rats but impaired it in spontaneously hypertensive rats. This suggests differential control in normotensive versus hypertensive rats. While we report data in normotensive humans in the present study that appear to be consistent with this observation, we do not have data on hypertensive humans. With regard to dietary salt loading studies (both animal and human), an important aspect may be the change or lack of...
change in plasma osmolality/sodium. That is, if excess dietary salt is consumed with an adequate amount of water intake, causing a “volume load” but no change in plasma osmolality (analogous to an acute ISO), then perhaps a decline in resting and baroreflex control of sympathetic activity would be predicted. If, on the other hand, dietary salt loading occurs without an adequate amount of water intake, causing an acute increase in plasma osmolality (analogous to a HSI), then perhaps an increase in resting and baroreflex control of sympathetic activity would be predicted (mechanisms underlying “thirst” responses may be relevant here). While this is clearly speculative, recent studies and reviews do emphasize the association of dietary salt intake and plasma sodium/osmolality (18) as well as an association between plasma sodium/osmolality and BP (2). Here, we simply suggest that salt intake has to be considered in relation to the water intake, and the resulting change or lack of change in plasma osmolality will have an affect on sympathetic outflow and control. Additional work to tease out these mechanisms is warranted.

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