Role of the paraventricular nucleus in renal excretory responses to acute volume expansion: role of nitric oxide

Yi-Fan Li, William G. Mayhan, and Kaushik P. Patel
Department of Physiology and Biophysics, University of Nebraska Medical Center, Omaha, Nebraska 68198-4575

Submitted 20 August 2002; accepted in final form 30 May 2003

Li, Yi-Fan, William G. Mayhan, and Kaushik P. Patel. Role of the paraventricular nucleus in renal excretory responses to acute volume expansion: role of nitric oxide. Am J Physiol Heart Circ Physiol 285: H1738–H1746, 2003; 10.1152/ajpheart.00727.2002.—Acute volume expansion (VE) produces a suppression of renal sympathetic nerve discharge (RSND) resulting in diuresis and natriuresis. Recently, we have demonstrated that the endogenous nitric oxide (NO) system within the paraventricular nucleus (PVN) produces a decrease in RSND. We hypothesized that endogenous NO in the PVN is involved in the suppression of RSND leading to diuretic and natriuretic responses to acute VE. To test this hypothesis, we first measured the VE-induced increase in renal sodium excretion and urine flow with and without blockade of NO, with microinjection of l-NAME; 200 pmol in 200 nl, within the PVN of Inactin-anesthetized male Sprague-Dawley rats. Acute VE produced significant increases in urine flow and sodium excretion, which were diminished in rats treated with l-NAME within the PVN. This effect of NO blockade within the PVN on VE-induced diuresis and natriuresis was abolished by renal denervation. Consistent with these data, acute VE induced a decrease in RSND (52% of the baseline level), which was significantly blunted by prior administration of l-NAME into the PVN (28% of the baseline level) induced by a comparable level of acute VE. Using the push-pull perfusion technique, we found that acute VE induced a significant increase in NOx concentration in the perfusate from the PVN region. Taken together, these results suggest that acute VE induces an increase in NO production within the PVN that leads to renal sympathoinhibition, resulting in diuresis and natriuresis. We conclude that NO within the PVN plays an important role in regulation of sodium and water excretions in the volume reflex via modulating renal sympathetic outflow.

renal sympathetic nerves; sodium retention

VOLUME CHALLENGES, such as the change in circulating blood volume, especially changes in arterial, atrial, and ventricular filling pressures, evoke reflex responses to regulate water and sodium excretion/retention by the kidney (7, 27). A decrease in circulating volume, such as an acute hemorrhage, induces a reflex increase in renal sympathetic nerve activity (1, 36) and the secretion of arginine vasopressin (AVP) (1, 26), resulting in an increase in the reabsorption of sodium and water in the kidney and a decrease in urine production. This reflex response prevents a decrease in body fluids to maintain blood pressure (BP). In contrast, when the circulating volume is increased, such as in experimentally induced volume expansion (VE), the reflex will develop in the opposite direction, inducing a decrease in renal sympathetic nerve activity and AVP secretion, resulting in an increase in sodium and water excretion (3, 14). This volume reflex plays an important role in balancing body fluid metabolism and stabilizing BP. The impairment of the volume reflex is believed to contribute to cardiovascular complications and aggravation of some disease states, including heart failure, hypertension, and diabetes (27, 29, 39, 43).

Sympathetic innervation in the kidney elicits important effects on the regulation of renal blood flow, renin release, and sodium and water reabsorption (6, 31). Thus reflex changes in renal sympathetic nerve activity to acute volume expansion play an important role in the efferent limb of the volume reflex (30). However, the details of the central nervous system pathway and neurotransmitter substances of this reflex arc are still not well established. The paraventricular nucleus (PVN) of the hypothalamus has been well known as an important integrating site in the forebrain to regulate sympathetic nerve activity (35). This, combined with the known role of the PVN in fluid balance and vasopressin release, puts the PVN in an unique position of regulating sodium excretion via renal nerves and water excretion via vasopressin release at rest and during volume challenges. Other studies (10, 19, 20), including our own, suggest that PVN neurons are involved in the mediation of the neural component of cardiovascular reflexes by influencing renal sympathetic nerve discharge (RSND). Furthermore, electrophysiological evidence shows that VE activates neurons within the PVN (2), indicating that the PVN is involved in the VE reflex response.

There is increasing evidence that nitric oxide (NO) acts as a nonsynaptic gaseous neurotransmitter to affect synaptic function in the central nervous system (4, 34). In the hypothalamus, NO synthase (NOS)-positive neurons are found primarily in the PVN and supraoptic nucleus (4, 23, 37). Studies from this and...
other laboratories (13, 40) have shown that the administration of an NO donor into the PVN produces a decrease in RSND, BP, and heart rate (HR) in rats. Conversely, the administration of a NO blocker [Nω-monomethyl-L-arginine (L-NMMA)] into the PVN increased RSND, BP, and HR (40).

On the basis of these facts, we hypothesized that NO within the PVN is involved in mediating changes in renal function in response to acute VE via regulating renal sympathetic nerve activity. The purpose of this study was to investigate the role of NO within the PVN in the renal excretory responses mediated by changes in renal sympathetic nerve activity to acute VE. To accomplish this, we tested 1) if blockade of NO in the PVN diminishes the VE-induced increase in renal sodium excretion and urine flow, 2) if this effect is dependent on intact renal sympathetic nerves, 3) if the blockade of NO in the PVN blunts the VE-induced RSND suppression, and 4) if VE induces an increase in NO release in the PVN.

METHODS

We used male Sprague-Dawley rats (250–300 g) obtained from Sasco Breeding Laboratories (Omaha, NE). All of the procedures used in this study were approved by the University of Nebraska Medical Center Institutional Animal Care and Use Committee. The experiments were conducted according to the "Guiding Principles for Research Involving Animals and Human Beings" of the American Physiological Society and National Institutes of Health regulations on experimental animals.

General Surgical Preparation

On the day of the experiment, the rat was anesthetized with urethane (0.75 g/kg ip) and α-chloralose (70 mg/kg ip) for renal nerve recording experiments and Inactin (100 mg/kg body wt) for renal function experiments. The left femoral vein was catheterized with polyethylene (PE) tubing (PE-50, filled with saline) for VE. The right femoral artery was cannulated with PE tubing (PE-50, filled with 100 U/ml heparinized saline) for arterial BP and HR monitoring. Central venous pressure (CVP) was measured by a catheter (PE-50) placed just outside the right atrium through the right jugular vein. All of the pressure signals, via pressure transducers (P231D, Gould), were fed to a computer-run data recording and analyzing system (PowerLab, ADInstruments). The trachea was intubated to facilitate ventilation.

Placement of Microinjection and Push-Pull Perfusion Cannulae Into the PVN

The anesthetized rat was placed in a stereotaxic apparatus (Davis Kopf Instruments; Tujunga, CA). A longitudinal incision was made on the head, and the bregma was exposed. The coordinates for the PVN were determined from the Paxinos and Watson Atlas (32) (1.5 mm posterior to the bregma, 0.4 mm lateral to the midline, and 7.8 mm ventral to the dura). A small burr hole was made in the skull. For the microinjections, a thin needle (0.5 mm outer diameter (OD) and 0.1 mm inner diameter (ID)) connected to a microsyringe (0.5 μl, model 7000.5, Hamilton microsyringe) was lowered into the PVN. For push-pull perfusion, a probe (0.7 mm OD and 0.2 mm ID) was lowered, which guided the inner (push) cannula into the PVN and collected the perfusate into the outer (pull) cannula. Both the push and pull cannulae were connected to a pump, which infused and returned artificial cerebrospinal fluid [aCSF; composition (in mM) 145 NaCl, 3.5 KCl, 1.0 MgCl₂, and 1.3 CaCl₂; pH 7.2] at a constant flow rate of 2–3 μl/min. The returned perfusate was collected as a fraction every 20 min. The samples were rapidly frozen at −70°C for nitrate/nitrite (NOₓ) measurement.

Volume Expansion

A continuous perfusion of 0.9% NaCl solution via the catheter in the right femoral vein was used as the VE stimulation. The total perfusion volume was 10% body weight, which was infused at a constant flow rate over a 40-min period.

Urine Collection and Measurement of Urine and Sodium Excretion

The ureters were catheterized with PE-10 tubing for collection of urine. The urine was collected before, during, and after VE, and the urine volume of each period was measured. The data are expressed as microliters per minute per gram kidney weight. Sodium concentration in the urine of each fraction was measured using an Ion/pH dual meter with a probe for sodium detection, allowing the probe to equilibrate in the sample for 2 min before a reading was obtained. The data are expressed as microequivalents per minute per gram kidney weight.

Renal Sympathetic Nerve Denervation

Both kidneys were exposed, and the renal sympathetic nerve trunks were isolated and destroyed using 95% ethanol. This method of renal denervation has been shown to decrease norepinephrine levels in the kidneys to an undetectable range (28).

Recording RSND

The left kidney was exposed through a retroperitoneal flank incision, and a branch of the renal nerve was isolated from the fat and connective tissue. The nerve was placed on a pair of thin bipolar platinum electrodes. The nerve-electrode junction was electrically insulated from the surrounding tissue with silicone gel (604 A and B, Wacker Sil-Gel). The electrical signal was amplified (10,000 times) with a Grass amplifier (P55) with a high- and low-frequency cutoff of 1,000 and 100 Hz, respectively. The output signal from the Grass amplifier was directed to a computer-run data-acquisition system (PowerLab) to record and integrate the raw nerve discharge. The signal recorded at the end of the experiment (after the rat was dead) was deemed as the background noise level. During the experiment, the value of the nerve discharge was calculated by subtracting the background noise value from the actual recorded value. The basal value of the nerve discharge was defined by subtracting the background noise value from the actual nerve discharge value before the administration of drugs into the PVN. The response of RSND to acute VE during the experiment (averaged over a period of 1 min) was subsequently expressed as the percent change from baseline.

Measurement of NO Release in the Perfusates From the PVN

NO in the samples of perfusate drawn from the PVN was measured via its NOₓ metabolites using a chemilumines-
cience detector (model 280, Sievers Nitric Oxide Analyzer). A standard curve for NaNO₃ concentration (100 μL of 0.1, 0.5, 1, 2.5, 5, and 10 μM) was generated for each experiment, and unknown samples were compared with the standard curve using the software provided with the Sievers Nitric Oxide Analyzer. This program takes into account both the peak response and the total area of the curve generated by standard and unknown samples. All measurements were performed, at least, in duplicate and then averaged to represent the mean for each sample.

**Brain Histology**

After the experiment, the rat was killed, and the brain was removed and fixed in 10% formalin for at least 24 h. The brain was then frozen, and serial transverse sections (30 μm) were cut with a cryostat (−18°C). The sections were mounted on microscope slides and then stained using 1% neutral red. The location of the injection within the PVN was verified under a microscope with ×40 magnification. Those injections with termination in the boundaries of the PVN were considered to be effective injections.

**Experimental Protocols**

**Experiment 1: effect of blockade of NO synthesis in the PVN on VE-induced changes in sodium excretion and urine flow in intact rats.** In the rats of the L-NMMA-treated group, 200 pmol of L-NMMA in 200 nl were injected into the PVN (unilateral injection) over a 10-min period at the beginning of the VE (L-NMMA and VE were begun simultaneously). In the control group, 200 nl of aCSF were injected into the PVN instead of L-NMMA. The urine produced was consecutively collected fractionally before (baseline) and during VE application. The volumes of urine produced and sodium concentration were measured afterward.

**Experiment 2: effect of blockade of NO synthesis in the PVN on VE-induced changes in sodium excretion and urine flow in intact rats.** In the rats of the L-NMMA-treated group, 200 pmol of L-NMMA in 200 nl were injected into the PVN over a 10-min period at the beginning of the VE process. In the control group, 200 nl of aCSF were injected into the PVN instead of L-NMMA. The urine product was consecutively collected before (baseline) and during VE application. The volumes of urine produced and sodium concentration were measured afterward.

**Experiment 3: effect of blockade of NO synthesis in the PVN on VE-induced changes in RSND.** To determine the effect of blockade of NO synthesis in the PVN on VE-induced changes in RSND, rats were treated with either L-NMMA (at the beginning of VE application, 200 pmol of L-NMMA in 200 nl were injected into the PVN over a 10-min period) or aCSF (200 nl) into the PVN (unilateral injection). BP, HR, CVP, and RSND were consecutively monitored before (baseline) and during VE.

**Experiment 4: change in the NOx level in the PVN during acute VE.** To determine the change in the NO level in the PVN during VE, a push-pull technique was used on anesthetized rats. Perfusates from the PVN were collected before (baseline, one 20-min collection) and during VE (two 20-min collections). Each fraction was analyzed for NOx separately by a NO analyzer.

**Data Analysis**

Responses of RSND to acute VE were expressed as the percent change from the baseline value. The basal value and the value after acute VE were subjected to two-way repeated-measures ANOVA, followed by comparison for individual differences using the Newman-Keuls test (39). Responses of arterial BP and HR to the drugs were expressed as the difference between the baseline and maximum response. *P* < 0.05 was considered to indicate statistical significance. All data are presented as means ± SE.

**RESULTS**

**General Values**

Figure 1 illustrates the histological data of the termination site of microinjection and push-pull perfusion
into the PVN. Among the 44 effective injection sites, 6 sites were used for experiment 1, 14 sites were used for experiment 2, and 12 sites each were used for experiments 3 and 4. The basal values (before VE or L-NMMA treatment) of body weight, BP, HR, RSND, urine flow, and sodium excretion in the rats of each experimental group are summarized in Table 1. There was no significant difference in all baseline levels of the observed parameters between the control and treated groups in any of the experiments. Arterial BP and HR were monitored during VE, and no significant change in BP or HR was found between the groups during acute VE (Table 2).

**Experiment 1: Effect of Blockade of NO Synthesis in the PVN on VE-Induced Changes in Sodium Excretion and Urine Flow in Intact Rats**

During the VE, urine flow was gradually increased in the normal control group, reaching $87 \pm 5 \mu\text{l} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$ at the highest response level. This increase in urine flow in response to VE was significantly attenuated in the L-NMMA-treated group compared with the control group, reaching only $59 \pm 11 \mu\text{l} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$ at the highest response level (Fig. 2A). Also, as shown in Fig. 2B, during VE, the response of sodium excretion was gradually increased in the control group, reaching $11 \pm 3 \mu\text{eq} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$ at the highest response level. However, this increase in sodium excretion was significantly diminished in the L-NMMA-treated group, reaching only $5 \pm 1 \mu\text{eq} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$ at the highest response level. Significant changes are observed with fairly modest changes in VE (2.5% body weight) as well. In the few cases where the microinjections were outside the boundaries of the PVN (>0.5 mm), L-NMMA injections were ineffective in producing any significant changes in normal sodium excretion to VE.

In normal rats without acute VE, blockade of NO with L-NMMA within the PVN produced a slight but significant increase in BP and HR (Fig. 3, C and D). However, L-NMMA within the PVN did not induce a significant change in urine flow or sodium excretion compared with the control rats with CSF injection within the PVN (Fig. 3, A and B).

**Experiment 2: Effect of Blockade of NO Synthesis in the PVN on VE-Induced Changes in Sodium Excretion and Urine Flow in Renal Sympathetic Denervated Rats**

In renal sympathetic denervated rats, VE-induced increases in urine flow and sodium excretion were still present and slightly elevated, reaching $100 \pm 7 \mu\text{l} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$ and $13.7 \pm 1.2 \mu\text{eq} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$, respectively, at the highest response level. The administration of L-NMMA into the PVN failed to significantly attenuate these diuretic and natriuretic responses, reaching $95 \pm 7 \mu\text{l} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$ and $11 \pm 1 \mu\text{eq} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$, respectively, at the highest response level, as observed in intact rats induced by VE (Fig. 4).

**Experiment 3: Effect of Blockade of NO Synthesis in the PVN on VE-Induced Changes in RSND**

As shown in Fig. 5A, in control rats, RSND was gradually decreased during acute VE, reaching $48 \pm 6\%$ of the baseline level at the highest response. In L-NMMA-injected rats, however, the response of RSND to VE was significantly attenuated, retaining $72 \pm 8\%$ of the baseline level at the highest response. Figure 5B shows that the alteration of RSND induced by VE was CVP associated. Similarly, the response of RSND versus CVP to VE in the L-NMMA-treated group was significantly diminished. Significant changes are observed with fairly modest changes in VE (2.5% body wt) as well. In the few cases where the microinjections were outside the boundaries of the PVN (>0.5 mm), L-NMMA injections were ineffective in producing any significant changes in normal sympathoinhibition to VE.

**Experiment 4: Change in the NO$_x$ Level in the PVN During Acute VE**

The mean data of baseline fractions (20 min) and the first fractions (20 min) during VE are shown in Fig. 6.

---

### Table 1. Basal values in various groups

<table>
<thead>
<tr>
<th>Experiment</th>
<th>n</th>
<th>BW, g</th>
<th>BP, mmHg</th>
<th>HR, beats/min</th>
<th>RSND, V·s</th>
<th>Urine Flow, $\mu\text{l} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$</th>
<th>Urinary Na$^+$ Excretion, $\mu\text{eq} \cdot \text{min}^{-1} \cdot \text{g kidney wt}^{-1}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>6</td>
<td>262 ± 12</td>
<td>101 ± 6</td>
<td>355 ± 23</td>
<td>2.6 ± 0.6</td>
<td>0.06 ± 0.01</td>
<td></td>
</tr>
<tr>
<td>L-NMMA</td>
<td>6</td>
<td>271 ± 18</td>
<td>110 ± 5</td>
<td>361 ± 21</td>
<td>2.4 ± 0.4</td>
<td>0.05 ± 0.01</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>6</td>
<td>268 ± 15</td>
<td>98 ± 9</td>
<td>354 ± 24</td>
<td>3.6 ± 0.7</td>
<td>0.1 ± 0.03</td>
<td></td>
</tr>
<tr>
<td>L-NMMA</td>
<td>6</td>
<td>267 ± 16</td>
<td>100 ± 9</td>
<td>351 ± 22</td>
<td>4.0 ± 0.6</td>
<td>0.09 ± 0.04</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>7</td>
<td>264 ± 14</td>
<td>104 ± 8</td>
<td>364 ± 26</td>
<td>5.6 ± 1.2</td>
<td></td>
<td></td>
</tr>
<tr>
<td>L-NMMA</td>
<td>7</td>
<td>259 ± 12</td>
<td>101 ± 6</td>
<td>357 ± 22</td>
<td>5.8 ± 1.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>6</td>
<td>261 ± 14</td>
<td>104 ± 5</td>
<td>354 ± 22</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE; n, no. of animals. BW, body weight; BP, blood pressure; HR, heart rate; RSND, renal sympathetic nerve discharge; L-NMMA, $N^\omega$-monomethyl-L-arginine.
During the period of VE, NO\textsubscript{x} concentration in the push-pull perfusate was significantly increased compared with that of the baseline. The value of NO\textsubscript{x} concentration in the fraction of the second 20 min of VE was slightly lower than in the first fraction but still significantly higher than the baseline. The results indicate that acute VE induced an increase in NO concentration in the PVN.

**DISCUSSION**

In the present study, we observed that acute VE-induced CVP-associated diuresis and natriuresis were blunted by the blockade of NO within the PVN. This effect is mediated via the renal sympathetic nerves, because renal denervation abolished the attenuation. Direct measurement of renal nerve activity indicated a blunted renal sympathoinhibition induced by VE after the central administration of an NO blocker within the PVN. Furthermore, we observed that acute VE induced an increase in NO\textsubscript{x} in the perfusate of the PVN. Taken together, these data indicate that NO within the PVN is involved in renal sympathoinhibition during acute VE.

NO, as an atypical gaseous neuromodulator, elicits multiple effects on the central nervous system by regulating presynaptic neurotransmitter transport and release as well as postsynaptic receptor activity (4, 15, 22). Neuronal NO plays an important role in regulating sympathetic nerve activity (16, 38). In many cases, NO elicits a so-called “breaking effect,” i.e., NO reduces neuronal activity to prevent overexcitation. Our previous study (40) has shown that the administration of an NO donor into the PVN produces a decrease in RSND, suggesting that NO in the PVN is involved in the inhibitory regulation of renal sympathetic activity. Our present study indicates that this inhibitory action of NO in the PVN plays a role in the reflex regulation of sympathetic activity in the volume-overloaded condition.

NO is also synthesized in the kidneys and influences renal functions (24), and the peripheral effect of L-NMMA must also be considered. However, the amounts of L-NMMA in the present study within the PVN were very small (200 pmol at the highest dose), hardly eliciting any observable effect directly on the kidney. Moreover, our denervation experiment demonstrated that the effect of administration of L-NMMA within the PVN was mainly via the renal sympathetic nerves.

Sympathetic nerve terminals have been identified in the renal vessels, the tubules, and the juxtaglomerular...
granular cells of the kidney. These sympathetic nerves regulate the major functions of the kidneys, including water and sodium excretion and reabsorption, renin secretion, and renal blood flow (9, 12). Thus VE-induced decreases in renal sympathetic activity result in an increase in the excretion of sodium and water. In the present study, it was found that blocking NO synthesis with L-NMMA in the PVN blunted the decrease in RSND and diminished the VE-induced diuretic and natriuretic responses. Moreover, we found that renal denervation abolished the effect of L-NMMA on the VE-induced altered renal functions, further suggesting that the action of NO in the PVN on renal function was mediated via alterations in renal sympathetic nerve activity. On the basis of the results of the present study, we conclude that NO in the PVN is involved in the mechanism of volume loading-induced reflex regulation of sodium and water metabolism in the kidney. These data are consistent with the view that VE stimulation induced an increase in NO synthesis and release in the PVN. NO then inhibited the sympathetic nervous system-associated neurons in the PVN and resulted in the decrease in sympathetic outflow, which then resulted in decreases in sodium and water reabsorption.

In normal rats without VE, however, administration of L-NMMA into the PVN did not induce significant changes in urine flow or sodium excretion, suggesting that blockade of NO within the PVN does not elicit

Fig. 3. Line plots showing the changes in urine flow rates (A), sodium excretion rates (B), blood pressure (BP; C), and and heart rate [HR, in beats/min (bpm); D] in anesthetized rats without volume expansion. ○, Mean values of the control group (injection of artificial cerebrospinal fluid (CSF), n = 6); ●, mean values of the group with L-NMMA administration into the PVN (n = 6).

Fig. 4. Line plots showing the changes in urine flow rates (A) and sodium excretion rates (B) of anesthetized bilateral renal sympathetic denervated rats undergoing volume expansion. ○, Mean value of the control group (n = 6); ●, mean values of the group with L-NMMA administration into the PVN (n = 6).
significant effects on renal excretory functions. In our previous study (41), it was observed that blockade of NO within the PVN induced a marked attenuation of the decrease in RSND and a significantly blunted increase in urine production and sodium excretion caused by VE.

How peripheral VE can induce NO synthesis in the PVN remains to be addressed. It was reported that the major sensors of a volume challenge are the cardiopulmonary mechanical receptors, which are primarily located on the cardiac atrium and ventricle (3). Stimulation these receptors via increasing atrial pressure by volume loading induces the activation of neurons in the nucleus tractus solitarii (NTS) as well as the inhibition of renal sympathetic nerve activity (3). The anatomic evidence has shown that there is a direct projection from the NTS to the hypothalamus and other forebrain regions (33). Consistent with these observations, acute VE causes excitation of the majority of paraventricular splatal neurons in the PVN (19). Furthermore, both renal sympathoinhibitory responses (10) as well renal vasodilatory responses (20) to acute volume load are attenuated after lesions of parvocellular neurons in the PVN. These parvocellular neurons in the PVN contain neuronal NOS. Thus this is a possible pathway for the stimulation of NO synthesis in the PVN. However, more direct evidence on the afferent arc of this volume reflex that induces the increase in NO synthesis is still needed.

Our and other studies have revealed that the VE reflex was impaired and blunted in some diseases, including chronic heart failure (8, 31), hypertension (29), and diabetic (30) conditions. This blunted volume reflex resulted in water and sodium retention and

![Fig. 5. Line plots showing the changes in renal sympathetic nerve discharge (RSND) of anesthetized rats undergoing volume expansion. A: graph of RSND vs. infusion volume; B: graph of RSND vs. central venous pressure. RSND was expressed as the percent change from the baseline level before the infusion. ●, Mean values of the control group (n = 7); ●, mean values of the group with L-NMMA administration into the PVN (n = 7). *P < 0.05 compared with the control group.](http://ajpheart.physiology.org/)

![Fig. 6. Bar graph showing the change in nitrite/nitrate (NOx) concentrations in the push-pull perfusates from the PVN. Open bar, mean value of the baseline; solid bar, mean value during acute VE. (n = 6). *P < 0.05 compared with baseline.](http://ajpheart.physiology.org/)
overloading in these diseases, which is a major factor in the development and aggravation of these diseases. Furthermore, a decreased NO level or neuronal NOS activity in the hypothalamus or the PVN has been found in heart failure (39, 42) and hypertension (11, 25). The deficiency of NO in the PVN or other hypothalamic regions may be the major cause of the blunted volume reflex observed in these diseases.

In conclusion, this study demonstrates that the central NO system within the PVN contributes to renal nerve-mediated changes in diuresis and natriuresis to acute VE and thus plays an important role in the regulation of the volume reflex.

The technical assistance of Hong Zheng and Phyllis Anding is greatly appreciated.

DISCLOSURES

This study was supported by National Heart, Lung, and Blood Institute Grant PO1-HL-62222.

REFERENCES


