Heme Oxygenase Inhibitor Restores Arteriolar Nitric Oxide Function in Dahl Rats

Fruzsina K. Johnson, William Durante, Kelly J. Peyton, Robert A. Johnson

Abstract—Vascular tissues express heme oxygenase (HO), which metabolizes heme to form carbon monoxide (CO). CO relaxes vascular smooth muscle but inhibits nitric oxide (NO) formation. Decreased NO synthesis may contribute to salt-induced hypertension in Dahl salt-sensitive (DS) rats. The current study examines the hypothesis that elevated levels of endogenous CO contribute to NO dysfunction in salt-induced hypertensive DS rats. Male DS rats were placed on high-salt (8% NaCl, HS) or low-salt (0.3% NaCl, LS) diets for 4 weeks. With respect to the LS group, the HS group's blood pressure and carboxyhemoglobin levels were elevated, and abdominal aortas showed 6-fold higher HO-1 protein levels. Experiments used isolated pressurized first-order gracilis muscle arterioles superfused with oxygenated modified Krebs buffer. An inhibitor of NO synthase, *N*-nitro-L-arginine methyl ester (L-NAME), caused concentration-dependent vasoconstriction in both groups, with attenuated responses in HS arterioles. HS arterioles also showed attenuated vasodilatory responses to an endothelium-dependent vasodilator, acetylcholine. Acute pretreatment with an inhibitor of HO, chromium mesoporphyrin, enhanced vascular responses to L-NAME and acetylcholine in both groups but abolished the differences between HS and LS arterioles. These data show that HO-1 protein levels and CO production are increased in HS rats. Arteriolar responses to L-NAME and acetylcholine are impaired in HS rats but abolished the differences between HS and LS arterioles. These results suggest that elevated levels of endogenous CO contribute to arteriolar NO dysfunction in DS rats with salt-induced hypertension. *(Hypertension. 2003;41:149-155.)*

Key Words: acetylcholine ▪ nitric oxide ▪ hypertension, sodium-dependent ▪ rats, Dahl ▪ endothelium

Carbon monoxide can be generated in the body mainly through the enzymatic degradation of heme by heme oxygenase.¹ Numerous tissues,² including vascular endothelial and smooth muscle cells, express heme oxygenase.³,⁴ The 2 major active isoforms of heme oxygenase are the inducible heme oxygenase-1 and the constitutive heme oxygenase-2. Pathological conditions,² such as angiotensin II–induced hypertension,⁵ can increase heme oxygenase-1 expression. There is now considerable evidence to suggest that endogenous carbon monoxide participates in the regulation of cardiovascular functions.⁶,⁷ Although carbon monoxide relaxes vascular smooth muscle,⁸ it also has been shown to interfere with the vasodilatory effects of the nitric oxide system.⁹–¹¹ We previously found that heme-derived carbon monoxide promoted endothelium-dependent and nitric oxide synthase–dependent vasoconstriction of skeletal muscle arterioles isolated from male Sprague-Dawley rats.¹²,¹³ These data suggested that carbon monoxide, even that which is endogenously formed, may attenuate nitric oxide function in normotensive animals. Because heme oxygenase can be induced by a variety of pathological conditions that are also associated with endothelial dysfunction, we were interested in identifying a pathological condition in which increased endogenous production of carbon monoxide may contribute to vascular nitric oxide dysfunction.

The Dahl/Rapp rats are either sensitive (DS) or resistant (DR) to the hypertensive effects of high salt diet.¹⁴ The DS rat is a genetic model of salt-induced hypertension because it develops hypertension on high-salt diet but remains normotensive on low-salt diet.¹⁵ Blood vessels isolated from hypertensive DS rats display impaired endothelium-dependent vasodilation,¹⁵,¹⁶ and basal nitric oxide function is attenuated in the microcirculation of DS rats during established salt-induced hypertension.¹⁷ Decreased nitric oxide formation has been suggested to contribute to salt-induced hypertension in DS rats;¹⁸ however, the pathological basis remains uncertain. Although substrate levels for nitric oxide synthesis are normal in these animals,¹⁹ salt-induced hypertension can be prevented²⁰,²¹ and reversed²² by the administration of l-arginine. Carbon monoxide has been shown to inhibit nitric oxide synthesis,⁹–¹¹ and excess l-arginine levels were reported to decrease the affinity of carbon monoxide binding to nitric oxide synthase.¹⁰ Furthermore, carbon monoxide has...
been reported to enhance the development of salt-induced hypertension in DS rats.\textsuperscript{22}

On the basis of these findings, we forwarded a hypothesis that heme-derived formation of carbon monoxide is increased in DS rats with salt-induced hypertension and contributes to arteriolar nitric oxide dysfunction. To test this hypothesis, we conducted experiments with skeletal muscle arterioles taken from DR and DS rats after 4 weeks of high- or low-salt diets and examined the responses to an inhibitor of nitric oxide synthase and an endothelium-dependent vasodilator while in the presence or absence of an inhibitor of endogenous carbon monoxide production.

**Methods**

**Materials**

Chromium mesoporphyrin (CrMP) was purchased from Frontier Scientific. Thiobutabarbitarial sodium (Inactin), 3,3’-diaminobenzidine (DAB), hematoxylin solution, \textit{N}-nitro-L-arginine methyl ester (L-NNAME), sodium nitroprusside, and acetylcholine were obtained from Sigma Aldrich. All other drugs were purchased from Fisher Scientific. CrMP stock solution (15 mmol/L) was prepared in 50 mmol/L Na\textsubscript{2}CO\textsubscript{3} solution and diluted in modified Krebs buffer (15 mmol/L) immediately before use. Acetylcholine (10 mmol/L) and sodium nitroprusside (1 mmol/L) stock solutions were prepared in saline and diluted in modified Krebs buffer immediately before use. L-NNAME was dissolved in modified Krebs buffer immediately before use. The composition of modified Krebs buffer was (mmol/L) immediately before use. Acetylcholine (10 mmol/L) and sodium nitroprusside (1 mmol/L) stock solutions were prepared in saline and diluted in modified Krebs buffer immediately before use.

**Animals**

Male inbred Dahl/Rapp salt-resistant (DR) (SR/Jr, \textit{n}=20) and salt-sensitive (DS) (SS/Jr, \textit{n}=64) rats were purchased at 5 to 6 weeks of age (Harlan, Indianapolis, Ind) and had free access to high-salt (8% NaCl) or low-salt (0.3% NaCl) diets (Dyets Inc) and tap water for 4 weeks. All procedures were approved by the institutional animal care committee.

**Blood Pressure and Carboxyhemoglobin Measurements and Tissue Extractions**

On the day of the experiment, rats were weighed, anesthetized, with a single injection of Inactin (100 mg/kg for DS rats and 140 mg/kg for DR rats IP), and a carotid arterial catheter was implanted for acute determination of blood pressure and heart rate and for blood sample collections. The carotid catheter was connected to a pressure transducer (TSD 104A, Biopac Systems) coupled to a polygraph system (Biopac Systems) and a personal computer. After obtaining stable readings, 3 blood samples (100 to 150 \textmu L) were drawn in 5-minute intervals for measurement of carboxyhemoglobin (HbCO) levels (OSM3 carboxyhemoglobinometer, Radiometer America Inc). Animals were then heparinized (1000 U/kg IV), and the heart, left kidney, a segment of the abdominal aorta, and the gracilis anticus muscles were removed and placed into ice-cold modified Krebs buffer. Left kidney and heart wet weights were then determined.

**Heme Oxygenase-1 Protein Measurements**

Abdominal aortic segments were harvested, snap-frozen in liquid nitrogen, and stored at \textminus 70°C until analyzed. Heme oxygenase-1 protein expression was determined by Western blotting, as previously detailed.\textsuperscript{23}

**Heme Oxygenase Immunohistochemistry**

Gracilis anticus muscles were harvested and fixed overnight (10% formalin). Specimens were embedded in paraffin and sectioned. Immunohistochemical staining for heme oxygenase-1 and heme oxygenase-2 was performed by using the avidin-biotin method (Vectastain Elite ABC kit, Vector Laboratories). Sections were deparaffinized and hydrated, and endogenous peroxidase activity was quenched. After incubation with rabbit polyclonal antibodies (Stressgen Biotechnologies Corp) against heme oxygenase-1 (1:3000 dilution) and heme oxygenase-2 (1:500 dilution), or incubation with vehicle only for control slides, sections were treated with biotinylated anti-rabbit IgG antibody. After incubation with the ABC reagent, sections were developed with DAB solution and counterstained with hematoxylin. The presence of heme oxygenase-1 and heme oxygenase-2 immunoreactivity was indicated by a brown color.

**Isolated Microvessel Experiments**

Segments of first-order gracilis muscle arterioles were isolated by microdissection.\textsuperscript{24} Individual arteriolar segments were cannulated at both ends with glass micropipettes in a water-jacketed vessel chamber.\textsuperscript{24} The distal micropipette was connected to a stopcock and the proximal micropipette to a reservoir whose height was adjusted to 108.8 cm to achieve 80 mm Hg intraluminal pressure. The vessel chamber was continuously superfused with gassed buffer (14% O\textsubscript{2}/5% CO\textsubscript{2}/balanced with N\textsubscript{2}; 37°C) through a nonrecirculating system. For internal diameter measurements, the vessel chamber was mounted on the stage of a microscope that was fitted with a videocamera leading to a videocaliper and a TV-VCR. With this setup, a magnified image of the arteriolar segment was viewed on the monitor, and the internal diameter was measured by manually adjusting the white guides superimposed by the video caliper. After a 60-minute stabilization period, the heme oxygenase inhibitor, 15 \mu mol/L CrMP, or matched vehicle was included in the superfusion buffer 20 minutes before the experiment. This pretreatment regime was continued throughout the remainder of the experiment. After the pretreatment period, increasing concentrations of a nitric oxide synthase inhibitor, L-NNAME (1 \mu mol/L to 3 mmol/L), or an endothelium-dependent vasodilator, acetylcholine (1 mmol/L to 3 \mu mol/L), were tested. For some experiments, after the 60-minute stabilization period, vessels were pretreated with an inhibitor of nitric oxide synthase (1 mmol/L L-NNAME for 45 minutes) to minimize endogenous nitric oxide production. After L-NNAME treatment, the heme oxygenase inhibitor, 15 \mu mol/L CrMP, or matched vehicle was included in the superfusion buffer 20 minutes before the experiment. This pretreatment regime was continued throughout the remainder of the experiment. After the pretreatment period, increasing concentrations of an endothelium-independent vasodilator, sodium nitroprusside (1 mmol/L to 3 \mu mol/L) were tested.

**Statistics**

All data are expressed as mean±SEM. Vascular response data were analyzed by ANOVA with a statistical package (SYSTAT). When significant differences were observed, orthogonal contrasts were performed as a post hoc analysis.\textsuperscript{25} All other data were analyzed by \textit{t} tests. A value of \textit{P}<0.05 was considered statistically significant.

**Results**

**Blood Pressure and HbCO Measurements**

The Table summarizes mean arterial pressure, heart rate, HbCO, and body and organ weights for the 4 groups. After 4 weeks of a high-salt diet, mean arterial pressure was increased in DS rats compared with the low-salt DS group. In contrast, mean arterial pressure was not different between the low- and high-salt groups in DR rats. Heart rate was not different between DR low- and high-salt or DS low- and high-salt animals. Compared with low-salt diet controls, DS rats after 4 weeks of a high-salt diet had lower body weights but higher kidney and heart weights. In contrast, body and heart weights were not different between the high- and low-salt DR groups, but kidney weight was elevated in DR rats on a high-salt diet. However, kidney weight was signif-
General Characteristics of Dahl Salt-Resistant and Salt-Sensitive Rats on High- and Low-Salt Diets

<table>
<thead>
<tr>
<th></th>
<th>Low Salt</th>
<th>n</th>
<th>High Salt</th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Salt-resistant</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MAP, mm Hg</td>
<td>131±3</td>
<td>10</td>
<td>134±3</td>
<td>10</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>383±9</td>
<td>10</td>
<td>391±6</td>
<td>10</td>
</tr>
<tr>
<td>Body weight, g</td>
<td>304±11</td>
<td>10</td>
<td>293±5</td>
<td>10</td>
</tr>
<tr>
<td>Kidney weight, g</td>
<td>1.15±0.03</td>
<td>10</td>
<td>1.30±0.02*</td>
<td>10</td>
</tr>
<tr>
<td>Heart weight, g</td>
<td>1.03±0.05</td>
<td>10</td>
<td>1.00±0.02</td>
<td>10</td>
</tr>
<tr>
<td>HbCO, %</td>
<td>3.5±0.1</td>
<td>9</td>
<td>3.5±0.1</td>
<td>9</td>
</tr>
<tr>
<td><strong>Salt-sensitive</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MAP, mm Hg</td>
<td>128±1</td>
<td>32</td>
<td>181±3†§</td>
<td>29</td>
</tr>
<tr>
<td>Heart rate, bpm</td>
<td>418±6§</td>
<td>32</td>
<td>411±8</td>
<td>29</td>
</tr>
<tr>
<td>Body weight, g</td>
<td>327±4</td>
<td>33</td>
<td>299±7†</td>
<td>30</td>
</tr>
<tr>
<td>Kidney weight, g</td>
<td>1.15±0.02</td>
<td>31</td>
<td>1.74±0.03‡</td>
<td>30</td>
</tr>
<tr>
<td>Heart weight, g</td>
<td>1.32±0.07‡</td>
<td>29</td>
<td>1.70±0.06‡</td>
<td>28</td>
</tr>
<tr>
<td>HbCO, %</td>
<td>3.6±0.1</td>
<td>6</td>
<td>4.0±0.1†§</td>
<td>6</td>
</tr>
</tbody>
</table>

Values are mean±SEM. DRLS indicates Dahl salt-resistant rats after 4 weeks of low-salt (0.3% NaCl) diet; DRHS, Dahl salt-resistant rats after 4 weeks of high-salt (8% NaCl) diet; MAP, mean arterial pressure; HR, heart rate; and HbCO, carboxyhemoglobin level.

*P<0.05 Dahl salt-resistant high-salt vs low-salt group.
†P<0.05 Dahl salt-sensitive high-salt vs low-salt group.
‡P<0.05 Dahl salt-resistant low-salt vs Dahl salt-sensitive low-salt groups.
§P<0.05 Dahl salt-resistant high vs Dahl salt-sensitive high-salt groups.

An inhibitor of nitric oxide synthase, L-NAME (1 μmol/L to 3 mmol/L), promoted concentration-dependent increases in internal diameter of arterioles isolated from DS rats after 4 weeks of high- and low-salt diets. However, the L-NAME–induced vasoconstriction was attenuated in high-salt arterioles compared with the low-salt group (low salt: Δmax 117±9 μm; n=5 versus high salt: Δmax 107±5 μm; n=10; P<0.05) (Figure 6). In contrast to the DS animals, there was no statistically significant difference in responses to an endothelium-dependent vasodilator, acetylcholine (1 mmol/L to 3 μmol/L) between high- and low-salt DR rat arterioles in the absence (low salt: Δmax 97±5 μm; n=6 versus high salt: Δmax 92±7 μm; n=5) (Figure 6). Acute pretreatment with an inhibitor of endothelial nitric oxide synthase, CrMP (15 μmol/L), enhanced L-NAME–induced vasoconstriction in both groups but abolished the difference between high- and low-salt DS rat arterioles (low salt: Δmax 63±8 μm; n=6 versus high salt: Δmax 59±9 μm; n=7) (Figure 5). However, there was no statistically significant difference in responses to an endothelium-dependent vasodilator, sodium nitroprusside (1 mmol/L to 3 μmol/L) between high- and low-salt DR rat arterioles in the absence (low salt: Δmax 98±16 μm; n=5 versus high salt: Δmax 87±12 μm; n=4) (Figure 6) or in the presence of the heme oxygenase inhibitor CrMP (15 μmol/L) (low salt: Δmax 124±9 μm; n=5 versus high salt: Δmax 117±6 μm; n=5) (Figure 6).
Discussion

In this study we found that vascular heme oxygenase-1 expression and endogenous carbon monoxide production were increased in Dahl salt-sensitive (DS) rats with salt-induced hypertension but not in Dahl salt-resistant (DR) rats on a high-salt diet. Vessels taken from the DS salt-hypertensive animals displayed altered responses to manipulations of the nitric oxide system, but DR high-salt arterioles did not. Furthermore, acute in vitro treatment with an inhibitor of endogenous carbon monoxide production abolished the differences between the DS hypertensive and normoten-sive arterioles.

Dahl/Rapp rats are either sensitive (DS) or resistant (DR) to the hypertensive effects of a high-salt diet.14 DS rats are a genetic model of salt-induced hypertension, and DR are a commonly accepted control strain for the salt-sensitive trait.14 We found that after 4 weeks, DS but not DR rats on the high-salt diet had higher mean arterial pressures than low-salt diet controls. Vessels taken from the DS salt-hypertensive animals displayed altered responses to manipulations of the nitric oxide system, but DR high-salt arterioles did not. Furthermore, acute in vitro treatment with an inhibitor of endogenous carbon monoxide production abolished the differences between the DS hypertensive and normoten-sive arterioles.

Carbon monoxide is a vasoactive byproduct of heme oxygenase–catalyzed breakdown of heme.3,7 Carbon monox-
The major endogenous source of carbon monoxide production is the heme oxygenase-catalyzed enzymatic degradation of heme.\(^3\)\(^,\)\(^7\) Numerous tissues,\(^2\) including vascular endothelial and smooth muscle cells, express heme oxygenase.\(^3\)\(^,\)\(^4\) To date, 3 heme oxygenase isoforms have been described. Heme oxygenase-1 (heat shock protein 32) is the inducible isoform because its gene expression can be increased severalfold by various stimuli.\(^2\) Heme oxygenase-2 is the constitutive isoform because its expression is relatively constant.\(^2\) Little is known about heme oxygenase-3 except that it has negligible catalytic activity compared with the other 2 isoforms.\(^2\)\(^,\)\(^8\) Previous studies suggested that angiotensin II–induced hypertension increases cardiac,\(^2\)\(^9\) aortic,\(^5\) and renal\(^1\) expression of heme oxygenase-1. Our current data show that abdominal aortic segments isolated from DS but not DR rats after 4 weeks of a high-salt diet contain higher heme oxygenase-1 protein levels compared with the low-salt group. We also found that in first-order gracilis muscle arterioles (the vessels we use for functional studies) isolated from hypertensive DS rats, heme oxygenase-1 immunostaining was enhanced in both the endothelial and vascular smooth muscle cells compared with low-salt controls. These data suggest that salt-induced hypertension in DS rats is accompanied by increased endogenous carbon monoxide production.

Decreased nitric oxide production has been suggested to contribute to salt-induced hypertension in DS rats;\(^1\) however, the pathological basis remains uncertain. Blood vessels isolated from hypertensive DS rats display impaired endothelium-dependent vasodilation and increased responsiveness to vasoconstrictors. Although substrate levels for nitric oxide synthesis are normal in these animals,\(^1\) salt-induced hypertension can be prevented\(^2\)\(^,\)\(^2\)\(^1\) and reversed\(^2\)\(^,\)\(^1\) by the administration of L-arginine. Carbon monoxide has been shown to inhibit nitric oxide synthase,\(^9\)\(^,\)\(^1\)\(^1\) and excess L-arginine levels were reported to decrease the affinity of carbon monoxide binding to nitric oxide synthase.\(^1\)\(^0\) We previously found that exogenous\(^1\)\(^2\) as well as endogenously formed\(^1\)\(^3\) carbon mon-
oxide promoted vasoconstriction in skeletal muscle arterioles isolated from male Sprague-Dawley rats. This carbon monoxide–induced vasoconstriction was abolished by endothelial removal,12,13 by inhibition of nitric oxide synthase,12,13 or by pretreatment with L-arginine.31 These data suggested that carbon monoxide promotes endothelium-dependent vasoconstriction most likely by inhibition of nitric oxide synthesis. Furthermore, induction of heme oxygenase-1 has been shown to attenuate muscarinic agonist–induced nitric oxide release11 and vasorelaxation32 in isolated renal arteries. Our current study shows that skeletal muscle arterioles isolated from DS rats after 4 weeks of a high-salt diet show impaired vasoconstrictor responses to an inhibitor of nitric oxide synthase, L-NAME, and attenuated vasodilatory responses to an endothelium-dependent vasodilator, acetylcholine, compared with low-salt controls. In contrast, vasodilatory responses to a nitric oxide donor, sodium nitroprusside, were not different between high- and low-salt DS arterioles. These data suggest that arterioles isolated from DS rats with salt-induced hypertension show impaired basal as well as receptor-stimulated nitric oxide function, which are not consequences of attenuated nitric oxide effectiveness. Furthermore, acute in vitro pretreatment with an inhibitor of endogenous carbon monoxide production, CrMP, enhanced arteriolar responses to L-NAME and acetylcholine and diminished the differences between high- and low-salt arterioles. In contrast, vasodilatory responses to the endothelium-dependent vasodilator acetylcholine were not different between high- and low-salt DR rats in the absence or presence of the heme oxygenase inhibitor. Our HbCO and heme oxygenase-1 protein measurements indicate that endogenous carbon monoxide production is increased during salt-induced hypertension in DS rats but not in DR rats on a high-salt diet. Taken together, these data suggest that vascular carbon monoxide production is increased after 4 weeks of salt-induced hypertension in DS rats, and it may contribute to arteriolar nitric oxide dysfunction by inhibiting nitric oxide synthesis. Furthermore, this heme oxygenase–mediated endothelial dysfunction does not appear to be a consequence of high-salt diet per se but rather is due to the combination of salt sensitivity, high-salt diet, and/or hypertension.

Previous studies suggested that a high-salt diet alone may alter vascular endothelial function13 or attenuate acetylcholine-induced vasodilation.38 In this study, we did not find a statistically significant difference in acetylcholine responses between high- and low-salt DR arterioles. Our results are in agreement with previous observations in DR rats by others.15 The differences may be rat strain–specific or due to different vascular beds used for the studies.

We observed that the endothelium-dependent vasodilatory responses to acetylcholine in DS rats on a low-salt diet are substantially attenuated compared with DR rats on low- or high-salt diets. Our data are in agreement with a recent study suggesting that the salt-sensitive trait per se can promote vasodilatory dysfunction in DS rats.35 Neither HbCO levels nor vascular heme oxygenase-1 expression were different between DS rats on a low-salt diet and DR rats on low- or high-salt diets. However, acute in vitro pretreatment with an inhibitor of heme oxygenase enhanced acetylcholine-induced vasodilation in DS rats on a low-salt diet but not in DR rats. Because substrate availability is normally a rate-limiting step for heme oxygenase–derived carbon monoxide formation,36,37 the possibility exists that heme formation might be enhanced in DS rats even on low-salt diets, which may contribute to endothelial dysfunction.

We have also noted that in our isolated microvessel experiments, responses to the nitric oxide synthase inhibitor L-NAME were much less attenuated during salt-induced hypertension than vasodilatory responses to acetylcholine. One possible explanation is that whereas L-NAME only promotes vasoconstriction, acetylcholine causes endothelium-dependent vasodilation but also promotes vasoconstriction through direct effects on vascular smooth muscle cells. Thus, acetylcholine-induced responses are a consequence of opposing vasodilatory and vasoconstrictor effects. Normally, the vasodilatory effects of acetylcholine are dominant in isolated rat skeletal muscle arterioles harvested from normotensive rats. It is possible that in our experiments, abolished acetylcholine responses in hypertensive DS rats may contribute to arterial dysfunction being only as large as the opposing vasodilator. The other possible explanation is that since acetylcholine-induced vasodilation is suggested to involve other mechanisms besides nitric oxide release (eg, prostaglandins, EDHF),38 these other vasodilatory pathways may also be inhibited by endogenous carbon monoxide in arterioles isolated from hypertensive DS rats.7

Perspectives

Our results suggest that endogenous carbon monoxide production is increased in Dahl rats after 4 weeks of salt-induced hypertension and that it contributes to arteriolar nitric oxide dysfunction. Endothelial dysfunction has been suggested to promote cardiac hypertrophy and renal damage in hypertensive Dahl rats.39 Our studies may provide some additional insights into the pathology of endothelial dysfunction and consequently might contribute to the understanding of the pathology of end-organ damage during salt-induced hypertension. Furthermore, angiotensin II–induced hypertension has been shown to increase cardiovascular expression of heme oxygenase-1.5,29,30 Therefore, the possibility exists that this phenomenon also extends to other forms of salt-sensitive hypertension.

Acknowledgments

This work was supported by National Heart, Lung, and Blood Institute grants R01HL64577 (PI, Robert A. Johnson, PhD) and R01 HL59976 (PI, William Durante, PhD), by the National Institutes of Health, Center of Biomedical Research Excellence in Hypertension and Renal Biology grant P20 RR17659 (PI, L. Gabriel Navar, PhD; Investigator, Fruzsina K. Johnson, MD), by an American Heart Association Established Investigator Grant (PI, William Durante, PhD) and Southeast Affiliate postdoctoral fellowship 0020335B (PI, Fruzsina K. Johnson, MD), and by the Solvay Pharmaceuticals Hypertension Yearly Grants Program (PI, Fruzsina K. Johnson, MD).

References

Heme Oxygenase Inhibitor Restores Arteriolar Nitric Oxide Function in Dahl Rats
Fruzsina K. Johnson, William Durante, Kelly J. Peyton and Robert A. Johnson

*Hypertension.* 2003;41:149-155; originally published online December 2, 2002;
doi: 10.1161/01.HYP.0000046923.52222.58

*Hypertension* is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2002 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/41/1/149

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in *Hypertension* can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to *Hypertension* is online at:
http://hyper.ahajournals.org//subscriptions/