Sodium-Induced Cardiac Hypertrophy
Cardiac Sympathetic Activity Versus Volume Load

Nicholas G. Fields, Baoxue Yuan, and Frans H.H. Leenen

To investigate the possible contributions of cardiac volume overload and cardiac sympathetic hyperactivity in the effects of sodium on cardiac mass, we evaluated the effects of treatment with saline (1%) and deoxycorticosterone acetate + saline (DOCA/saline) for 10 days and 3 and 6 weeks on ventricular anatomy and intracardiac pressures. Sympathetic activity in the heart and other tissues was assessed at 10 days and 3 weeks by catecholamine turnover rates and tyrosine hydroxylase activity. Both saline and DOCA/saline produced concentric left ventricular (LV) hypertrophy. Right ventricular weight showed only small increases. Saline treatment did not affect LV end-systolic pressure, whereas DOCA/saline caused a moderate increase (to 159 mm Hg). Right atrial pressure was not affected by either treatment, whereas LV end-diastolic pressure increased but only after the development of LV hypertrophy. Both saline and DOCA/saline decreased LV norepinephrine concentration; only DOCA/saline decreased norepinephrine content per LV. However, neither treatment altered the norepinephrine turnover rate constant, the absolute turnover rate, or the tyrosine hydroxylase activity. The results demonstrate that increased saline intake or DOCA/saline produces concentric LV hypertrophy without any increase in blood pressure in the case of saline and with increases in LV filling pressure following rather than preceding the appearance of LV hypertrophy. The lack of an increase in LV norepinephrine turnover and tyrosine hydroxylase activity suggests that the hypertrophy is not mediated through increased cardiac neuronal sympathetic activity. (Circulation Research 1991;68:745–755)

Besides pressure and volume overload,1,2 increases in cardiac sympathetic activity or renin activity3–5 may cause hypertrophic responses of myocardial tissue. Recent studies suggest that sodium can modulate cardiac mass, too. Increased dietary sodium has been shown to increase heart weight without increasing blood pressure,6 and sodium restriction reduced heart weight in two-kidney, one-clip hypertensive rats without a concomitant decrease in blood pressure.7

At present, the type of structural changes produced by increased sodium intake or the mechanisms by which they occur are unknown. If effects on cardiac volume load play a major role, one may expect development of eccentric left ventricular (LV) hypertrophy (increased internal diameter with little change in wall thickness) rather than concentric LV hypertrophy (increased wall thickness with little change in internal diameter).1 No data are available in this regard. Sodium-induced changes in cardiac norepinephrine (NE) concentrations6,7 and receptor characteristics8 have led to the hypothesis that the effects of sodium are mediated through alterations in cardiac sympathetic activity.6,7 However, none of these parameters actually reflects cardiac sympathetic activity.

To characterize the actual structural changes in the heart caused by increased sodium intake and possible underlying mechanisms, we examined the effects of 1% saline intake on ventricular anatomy and on cardiac sympathetic activity and volume load. To define the type (eccentric or concentric) of hypertrophy produced, changes in both LV and right ventricular (RV) weight, LV wall thickness, and LV internal diameter were determined.8,9 Sympathetic activity was assessed by catecholamine turnover rates and tyrosine hydroxylase activity. Cardiac volume load was assessed by measurement of cardiac filling pressures. The changes in saline-treated rats were compared with those in deoxycorticosterone acetate + saline (DOCA/saline)–treated rats in which the factors of elevated blood pressure and possibly

From the Hypertension Unit, University of Ottawa Heart Institute, and Department of Medicine, University of Ottawa, Ottawa, Canada.

Supported by an operating grant from the Medical Research Council of Canada. N.G.F. and B.Y. were supported by fellowships from Sandoz Canada Inc. F.H.H.L. was supported by a career investigatorship from the Heart and Stroke Foundation of Ontario.

Address for correspondence: Frans H.H. Leenen, MD, PhD, FRCP(C), Hypertension Unit, University of Ottawa Heart Institute, 40 Ruskin Street, Ottawa, Ontario, Canada, K1Y 4E9.

Received June 9, 1989; accepted November 6, 1990.
more marked cardiac sympathetic activity are present in addition to high salt intake.\textsuperscript{10}

**Materials and Methods**

Male Wistar rats weighing 140–170 g (Charles River Breeding Laboratories, Montreal, Canada) were used. The rats were housed two per cage in a temperature-controlled room with a 12-hour light and dark cycle and had free access to food (Purina rat chow, 180 \mu mol sodium/g food) and water. All rats underwent a left nephrectomy with halothane−nitrous oxide−oxygen anesthesia, and after surgery they were randomized to the appropriate treatment group. Three experimental protocols were performed for assessment of 1) hemodynamics and ventricular anatomy after 10 days and 3 and 6 weeks of treatment, 2) catecholamine turnover and tyrosine hydroxylase activity after 10 days and 3 weeks of treatment, and 3) plasma catecholamines and the blood pressure response to sympathetic blockade by prazosin and hexamethonium after 3 weeks of treatment. In all of the experiments body weight and food and fluid intake were recorded once per week.

**Assessment of Hemodynamics and Ventricular Anatomy**

Thirty rats per treatment period were randomized to either the untreated, saline, or DOCA/saline groups (n=10/group). Final numbers of the rats per group may vary either because of death during the cannulation procedure or because of the inability to obtain measurements through clotted blood in the cannulas. Twenty-four hours after surgery, rats in the DOCA/saline group received the first of twice weekly injections of DOCA (25 mg/kg s.c. in 0.1 ml sesame oil/100 g body wt\textsuperscript{12}). Rats in the untreated and saline groups received injections of sesame oil only. NaCl (1%) was substituted for drinking water in the DOCA/saline and saline groups, and animals in the untreated group received distilled water. The rats were subjected to 10 days or 3 or 6 weeks of treatment.

At the end of the treatment period, rats were anesthetized using halothane−nitrous oxide−oxygen anesthesia, and polyethylene catheters (PE 50, Clay Adams, Becton Dickinson Labware, Lincoln Park, N.J.) filled with heparinized saline were placed into the right atrium via the right jugular vein, and the LV via the left carotid artery, for determination of right atrial pressure, LV end-systolic pressure (LVESP), and LV end-diastolic pressure (LVEDP). After a 3-hour recovery period (a length of time sufficient for recovery from anesthesia\textsuperscript{12}) the rats were put into partitioned cages and the catheters were connected to pressure transducers (Statham P23 ID, Statham-Gould, Oxnard, Calif.). The intracardiac pressures were recorded on a Model 7D multichannel chart recorder (Grass Instrument Co., Quincy, Mass.) in conscious, unrestrained rats after a 30-minute acclimatization period. LVESP and LVEDP were recorded at a setting of 50 mm Hg/cm and right atrial pressure at a setting of 5 mm Hg/cm. Paper speed was 100 mm/sec. Subsequently, plasma volume and blood volume were determined by the RISA−\textsuperscript{131}I technique as described previously.\textsuperscript{13} At the end of the experiment, the animals were killed for determination of LV and RV weights and ventricular anatomy in a transverse midlevel slice of the LV as described previously.\textsuperscript{8} The midlevel slice of the LV was obtained by two transverse cuts at one third and two thirds of the length. For determination of dry LV and RV weights, the RV and three LV slices were dried in an oven at 37°C for 24 hours (sufficiently long for complete evaporation of tissue water).

**Assessment of Sympathetic Activity**

**Catecholamine turnover.** A total of 108 rats were used. Half of the rats were tested after 10 days of treatment, and the remaining rats were tested after 3 weeks of treatment. For each treatment period the rats were randomized to the untreated, saline, or DOCA/saline groups (n=18/group). Catecholamine turnover was determined in the LV, RV, superior and large mesenteric arteries, and adrenal medulla. The catecholamine turnover rate was calculated from the decline with time of endogenous NE or epinephrine (adrenal medulla only) after inhibition of synthesis with DL-\textsuperscript{d}-methyl-\textsuperscript{L}-tyrosine methyl ester HCl (metyrosine).\textsuperscript{14} Metyrosine was administered at a dose of 300 mg/kg i.p. in 0.15 ml distilled water/100 g body wt.\textsuperscript{15}

Eighteen rats from each group were decapitated before or 4.5 or 9 hours after (n=6 at each time period) metyrosine administration, with the rats in the 9-hour group receiving a maintenance dose of 300 mg/kg 4.5 hours after the first injection. After decapitation the hearts were quickly removed, rinsed in cold saline, blotted dry, and dissected free of the atria and great vessels; the LV and RV were separated and placed on dry ice. Subsequently, the adrenal gland and the large and superior mesenteric arteries were removed, cleaned of contaminating tissue, and also placed in dry ice. The tissues were stored at −85°C until homogenization. The frozen ventricles were weighed, and 100 mg of each (cut from the apex) was homogenized in 2 ml acetic acid (0.25 M) containing glutathione (1.6 mM) and EGTA (0.65 mM) with a polytron homogenizer (Brinkmann Instruments, Inc., New York). The portion of the mesenteric artery collected was also weighed, and a 5-mg portion was homogenized as described above. By using a dissecting microscope, the adrenal medulla was dissected free of the cortex before homogenization. The homogenates were centrifuged at 5,000g for 20 minutes at 4°C and stored at −85°C until extraction of the catecholamines. The catecholamines were extracted by adsorption onto alumina and were assayed using high-performance liquid chromatography (HPLC) with electrochemical detection.\textsuperscript{8} The HPLC system consisted of a Model 510 pump and an automatic injection system (WISP Model 712) obtained from Waters Associates, Milford, Mass. The 5-\mu m C\textsubscript{18} column (220×4.6-mm i.d.,
RP-18) was obtained from Brownlee Laboratories, Santa Clara, Calif. The LC 17 electrochemical detector (Bioanalytical Systems, Inc., West Lafayette, Ind.) was used with a glassy carbon electrode. The detector was maintained at +0.65 V versus an Ag-AgCl reference electrode. The mobile phase consisted of 0.15 M monochloroacetic acid, 2 mM Na₂EDTA, 3.75 mM sodium 1-octane sulfonic acid in water plus 5% methanol. The flow rate was 1.0 ml/min. The turnover rate constant (k) was calculated from the rate of the decline of the logarithm of tissue catecholamine concentration. The turnover rate was calculated from the following equation: turnover rate = [NE] × k, where [NE] is the concentration in the tissue before inhibition of synthesis.14

**Tyrosine hydroxylase activity.** Tissues for tyrosine hydroxylase activity were obtained from the rats killed at time zero in the turnover studies. Tyrosine hydroxylase activity was assessed in the LV and adrenal medulla by using a modification of the HPLC method of Nagatsu et al.16 Sufficient amounts of the other tissues were not available for determination of tyrosine hydroxylase activity. The tissues were homogenized in phosphate buffer (2 mM) containing 2% Triton X-100. The LV (400–500 mg) was homogenized in 4 vol (1.6–2.0 ml) buffer and the weighed adrenal medulla in 1.5 ml buffer. The homogenates were centrifuged at 15,000g for 15 minutes, and the supernatants were stored at −85°C until use. The reaction mixture consisted of the following components (final concentration in parentheses): 20 µl of 1 M acetate buffer, pH 6.0 (0.2 M), 20 µl of 2 mM L-tyrosine in 0.01 M HCl (0.2 mM), 10 µl of 10 mM 6-methyl-5,6,7,8-tetrahydropterin (1 mM) in 1 M 2-mercaptoethanol (100 mM), 30 µl of phosphate buffer, 10 µl of 10 mM ferrous ammonium sulfate (1 mM), and water. For the blank incubation, D-tyrosine was used as a substrate instead of L-tyrosine, and 100 pmol L-dopa was added to another blank incubation as a standard. The samples were run in duplicate, and incubation was carried out at 37°C. Incubation time was 1 hour for the LV and 15 minutes for the adrenal medulla. These periods were found in preliminary experiments to be adequate to produce the maximum activity. The HPLC system and mobile phase used were similar to those described for the detection of NE and epinephrine in the turnover studies, with a flow rate of 0.6 ml/min. This gave retention times of 7.5 minutes for L-dopa and 16 minutes for α-methyl-dopa, the internal standard.

**Assessment of plasma catecholamines and the blood pressure response to sympathetic blockade.** Twenty-one rats (seven/group) underwent surgery and randomization as described above. At the end of the treatment period the rats were anesthetized as previously described, and polyethylene catheters (PE 50) filled with heparinized saline were placed into the left carotid artery and right jugular vein. The next morning, the rats were placed in test cages, carotid cannulas were connected to pressure transducers, and blood pressures were displayed on a Grass polygraph.

After a 30-minute acclimatization period, resting blood pressures and heart rates were determined from the blood pressure tracings. The cannulas were then removed from the transducers, and a blood sample (400 µl) was collected into chilled microcentrifuge tubes containing EGTA (0.25 M), glutathione (0.2 M), and heparin for radioenzymatic determination of plasma catecholamines.17 After this, the cannulas were reconnected, blood pressure was stabilized for 15 minutes, and prazosin was infused through the jugular cannula by an infusion pump (model 944, Harvard Apparatus, South Natick, Mass.). The infusion rate was adjusted to give cumulative doses of prazosin 1, 5, and 10 mg/kg, each dose being administered over 5 minutes. Blood pressure and heart rate were determined at the end of each infusion period. After the prazosin infusion a bolus dose of hexamethonium (30 mg/kg) was administered through the carotid cannula, and the blood pressure and heart rate were determined.

**Statistical analysis.** For the turnover data, the turnover rates were expressed as means with 95% confidence limits instead of SEM because the distribution was asymmetrical.18 All other results are expressed as mean ± SEM. The data were analyzed by one-way analysis of variance, and the Duncan multiple range test was used for post hoc analysis. Values of p < 0.05 were considered statistically significant.

**Results**

**Ventricular Anatomy**

**Ventricular weight.** Saline alone increased LV mass, with the LV weight/body weight ratio in the saline-treated group being significantly increased compared with that of the untreated group after 3 (+ 10%) and 6 (+ 19%) weeks of treatment (Figure 1). DOCA/saline increased LV mass within 10 days of treatment, the increase becoming more marked after longer treatment periods. The absolute LV weight (Table 1) exhibited a similar pattern, and all of the above significant differences were evident except for those after 10 days of treatment. LV dry and wet weights changed in parallel and the dry weight/wet weight ratios were not affected after 3 and 6 weeks of treatment (24 ± 2%, 25 ± 1%, and 23 ± 1% for control, saline, and DOCA/saline groups, respectively, after 6 weeks).

RV weight showed modest increases in relative (Figure 1) and absolute (Table 1) weight after prolonged treatment with DOCA/saline. Saline alone caused small, nonsignificant increases in RV weight (Figure 1, Table 1). The dry weight/wet weight ratio did not change (25 ± 1%, 24 ± 1%, and 25 ± 1% for control, saline, and DOCA/saline groups, respectively, after 6 weeks).

**LV wall thickness and dimensions.** Neither of the treatments affected LV internal diameter, but significant increases in wall thickness were observed in the saline- and DOCA/saline-treated groups compared with those of the untreated group after 3 and 6 weeks.
Pressures and weights in untreated, saline-treated, and deoxycorticosterone acetate+saline–treated rats after 10 days, 3 weeks, and 6 weeks of treatment. Each bar represents the mean±SEM. *p<0.05 vs. untreated rats; **p<0.05 vs. untreated and saline-treated rats. Open bars, untreated rats (n=6, 9, and 8 at 10 days, 3 weeks, and 6 weeks, respectively); hatched bars, saline-treated rats (n=7, 10, and 7 at 10 days, 3 weeks, and 6 weeks, respectively); solid bars, DOCA/saline-treated rats (n=9, 8, and 8 at 10 days, 3 weeks, and 6 weeks, respectively).

of treatment (Figure 2). The magnitude of the increase in wall thickness in the DOCA/saline group was greater than in the saline group, significant after 3 but not 6 weeks of treatment.

Intracardiac Pressures and Heart Rate

Saline treatment did not alter LVESP (Figure 3). In the DOCA/saline rats LVESP was significantly elevated compared with LVESP levels of the saline and untreated groups after 10 days of treatment and continued to rise, reaching a value of 159±8 mm Hg after 6 weeks of treatment.

No significant differences were observed in LVEDP among the three groups after 10 days of treatment (Figure 3). In saline-treated rats, LVEDP showed a significant increase after prolonged (6 weeks) treatment. In the DOCA/saline-treated rats, LVEDP was significantly increased compared with LVEDP levels of both the saline and untreated groups after 3 and 6 weeks of treatment.

No significant changes in right atrial pressure were observed between the groups at any of the treatment periods. Figure 3.

No differences in heart rate were observed between the groups after 10 days or 3 weeks of treatment. After 6 weeks, heart rate in the DOCA/saline group was decreased compared with that of both the control and saline groups (348±19 versus 391±11 and 397±13 beats/min, respectively, p<0.01).

Plasma Volume, Blood Volume, and Hematocrit

No significant differences in plasma or blood volume were observed between the groups after 10 days or 3 weeks of treatment (Table 2). After 6 weeks of treatment, plasma volume was significantly increased in the DOCA/saline-treated rats compared with volumes of the other groups. Blood volume showed a nonsignificant increase at this time. No changes in hematocrit were observed after 10 days or 3 weeks of treatment, but a significant decrease was observed in the DOCA/saline-treated rats compared with the untreated and saline-treated rats after 6 weeks of treatment.

Sympathetic Activity

Tissue catecholamines and turnover. For the LVs in which significant hypertrophy occurred, the absolute turnover rates (NE\textsubscript{oxk}×k) were calculated using both the total organ content and the concentration per gram of tissue to account for the decreases in NE content caused by hypertrophy (Figure 4, Tables 3 and 4).

LV NE concentrations expressed per gram of tissue decreased in both saline- and DOCA/saline-treated rats (Table 3), whereas NE levels expressed per total LV decreased significantly only for the DOCA/saline group (Table 3). Neither treatment significantly altered the rate of decline (k) of LV NE (Table 4). Saline treatment did not significantly alter the absolute NE turnover rate whether expressed per

<table>
<thead>
<tr>
<th>Treatment duration</th>
<th>10 days</th>
<th>3 weeks</th>
<th>6 weeks</th>
</tr>
</thead>
<tbody>
<tr>
<td>LV weight (mg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Untreated</td>
<td>566±26</td>
<td>561±15</td>
<td>797±22</td>
</tr>
<tr>
<td>Saline</td>
<td>563±16</td>
<td>729±17*</td>
<td>937±14*</td>
</tr>
<tr>
<td>DOCA/saline</td>
<td>620±15†</td>
<td>834±16*</td>
<td>996±17*</td>
</tr>
<tr>
<td>RV weight (mg)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Untreated</td>
<td>116±2</td>
<td>144±5</td>
<td>166±10</td>
</tr>
<tr>
<td>Saline</td>
<td>117±4</td>
<td>162±3†</td>
<td>183±11</td>
</tr>
<tr>
<td>DOCA/saline</td>
<td>120±3</td>
<td>166±11*</td>
<td>193±16</td>
</tr>
</tbody>
</table>

Values are mean±SEM (for n values, see legend to Figure 1). *p<0.05 vs. untreated rats. †p<0.05 vs. untreated saline-treated rats.
gram of tissue (Table 4) or per total LV (Figure 4). In the DOCA/saline group, the absolute turnover rate per gram of tissue was significantly decreased at both 10 days and 3 weeks of treatment (Table 4). However, the absolute turnover rate expressed per total LV was not significantly reduced (Figure 4).

In contrast to the LV, there was no difference in the NE concentration in the RV between the groups (Table 3). Saline alone did not change the rate of decline of NE or the absolute turnover rate in the RV. DOCA/saline caused small (p<0.1) increases after 3 weeks (Figure 4, Table 4).

In the superior and large mesenteric arteries, both saline and DOCA/saline decreased NE concentrations (Table 3). However, there were no differences in the rate of decline of NE between the groups. Because of the reduced NE concentrations, the absolute turnover rates in the treated groups were decreased compared with those of the untreated group (for significant decreases see Table 4).

Neither treatment significantly altered the catecholamine concentrations or the rate of decline and turnover rate of NE in the adrenal medulla (Tables 3 and 4). A significant increase in the turnover rate of epinephrine in the adrenal medulla was observed in the DOCA/saline group after 3 weeks of treatment (Table 4).

**Tyrosine hydroxylase activity.** Saline treatment alone did not affect tyrosine hydroxylase activity in either the LV or adrenal medulla (Figure 5). DOCA/saline caused small increases in tyrosine hydroxylase activity in the LV after 10 days (NS) and 3 weeks (p=0.06) of treatment. In the adrenal medulla...
DOCA/saline significantly increased tyrosine hydroxylase activity after 3 weeks.

**Plasma catecholamines.** Neither treatment altered plasma NE levels (Table 5). DOCA/saline treatment did, however, significantly increase plasma epinephrine levels.

**Blood pressure response to prazosin and hexamethonium.** The decreases in systolic and diastolic blood pressures caused by prazosin were significantly greater in the DOCA/saline group than in the other groups (Figure 6). There was no difference, however, in the hypotensive effect of prazosin between the saline and untreated groups. Following prazosin, hexamethonium caused only minor, nonsignificant further decrease in blood pressure, indicative of an adequate sympathetic blockade by prazosin. After prazosin (± hexamethonium), the blood pressure of the three treatment groups no longer differed significantly.

**Body Weight and Fluid Intake**

Body weight in the untreated and saline-treated rats increased steadily to about 400 g at the end of 6 weeks (Table 6). In DOCA/saline-treated rats, body weight increased at the same rate as in the untreated group for the first 3 weeks of treatment. After this, however, the DOCA/saline-treated rats did not gain weight, and after 6 weeks of treatment, body weight in this group was significantly decreased compared with the weights of the untreated and saline groups, a phenomenon described previously.19

Fluid intake was significantly higher in the saline and DOCA/saline groups compared with that of the untreated group (Table 6). The fluid intake in the DOCA/saline group was also higher than in the saline group.

**Discussion**

Two major new findings emerge from the present study. 1) Both saline and DOCA/saline cause concentric LV hypertrophy, in the case of saline without an increase in blood pressure, and not eccentric hyper-
TABLE 3. Catecholamine Concentrations in Untreated, Saline-Treated, and Deoxycorticosterone Acetate+Saline-Treated Rats

<table>
<thead>
<tr>
<th></th>
<th>Treatment duration</th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10 days</td>
<td>3 weeks</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Untreated</td>
<td>Saline</td>
<td>DOCA/saline</td>
</tr>
<tr>
<td><strong>Norepinephrine</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left ventricle</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ng/g</td>
<td>546±41</td>
<td>430±52</td>
<td>378±38*</td>
</tr>
<tr>
<td>ng/LV</td>
<td>224±26</td>
<td>200±30</td>
<td>193±10</td>
</tr>
<tr>
<td>Right ventricle</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ng/g</td>
<td>733±62</td>
<td>804±138</td>
<td>838±121</td>
</tr>
<tr>
<td>ng/RV</td>
<td>87±9</td>
<td>87±18</td>
<td>96±15</td>
</tr>
<tr>
<td>Superior mesenteric arteries ng/g</td>
<td>4,900±116</td>
<td>4,274±295</td>
<td>4,068±303τ</td>
</tr>
<tr>
<td>Large mesenteric arteries ng/g</td>
<td>15,295±1,198</td>
<td>11,574±814†</td>
<td>11,100±714†</td>
</tr>
<tr>
<td>Adrenal medulla</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>µg/g</td>
<td>3,832±161</td>
<td>4,137±157</td>
<td>4,054±482</td>
</tr>
<tr>
<td>Epinephrine content</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adrenal medulla</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>µg/g</td>
<td>18,865±948</td>
<td>16,252±691</td>
<td>18,353±2,510</td>
</tr>
</tbody>
</table>

Values are mean±SEM for six rats. DOCA/saline, deoxycorticosterone acetate+saline; LV, left ventricle; RV, right ventricle.
*p<0.05 vs. untreated and saline-treated rats.
†p<0.05 vs. untreated rats.

The development of cardiac hypertrophy, likely reflecting a decreased compliance of the hypertrophied ventricle rather than a cause of the hypertrophy.1 The lack of any evidence for eccentric hypertrophy was somewhat unexpected because saline intake could conceivably lead to fluid retention, and mineralocorticoid hypertension is often considered to be a "volume-dependent" hypertension.20,21 However, determination of cardiac filling pressures had not yet been performed, whereas increases in intravascular volume are not consistently present.20,22,23

The results of the present study also indicate that cardiac neuronal sympathetic activity does not increase during the development of saline- and DOCA/saline-induced cardiac hypertrophy. Cardiac sympathetic activity was assessed through measurement of catecholamine turnover rates and tyrosine hydroxylase activity, which provide independent assessments of the level of neuronal sympathetic activity.14,24,25 Consistent with other studies,6,26,27 saline and DOCA/saline treatment decreased NE concentration in the LV and mesenteric arteries. However, the NE concentration does not reflect actual sympathetic activity. Indeed there is no evidence for increased LV sympathetic neuronal activity from either the turnover or tyrosine hydroxylase data. In the turnover studies, neither the rate constant (k) of decline of NE, which is indicative of neuronal firing and reuptake, nor the absolute turnover rate was significantly increased in the LV or mesenteric arteries of either group compared with those of the untreated group.

In the absence of cardiac sympathetic neuronal hyperactivity, it is possible that cardiac sympathetic...
TABLE 4. Catecholamine Turnover Rates in Untreated, Saline-Treated, and Deoxycorticosterone Acetate+Saline-Treated Rats

<table>
<thead>
<tr>
<th></th>
<th>Untreated</th>
<th>Saline</th>
<th>DOCA/saline</th>
<th>Untreated</th>
<th>Saline</th>
<th>DOCA/saline</th>
<th>Untreated</th>
<th>Saline</th>
<th>DOCA/saline</th>
</tr>
</thead>
<tbody>
<tr>
<td>Left ventricle</td>
<td>-0.10±0.02</td>
<td>0.10±0.02</td>
<td>-0.08±0.02</td>
<td>-0.10±0.01</td>
<td>0.10±0.02</td>
<td>-0.03±0.02</td>
<td>-0.10±0.01</td>
<td>0.10±0.02</td>
<td>-0.03±0.02</td>
</tr>
<tr>
<td>10 days</td>
<td>k (hr⁻¹)</td>
<td>Turnover rate (ng/g/hr)</td>
<td>k (hr⁻¹)</td>
<td>Turnover rate (ng/g/hr)</td>
<td>k (hr⁻¹)</td>
<td>Turnover rate (ng/g/hr)</td>
<td>k (hr⁻¹)</td>
<td>Turnover rate (ng/g/hr)</td>
<td>k (hr⁻¹)</td>
</tr>
<tr>
<td>Right ventricle</td>
<td>-0.05±0.01</td>
<td>0.06±0.03</td>
<td>-0.05±0.02</td>
<td>-0.03±0.02</td>
<td>0.03±0.02</td>
<td>-0.06±0.02</td>
<td>-0.05±0.02</td>
<td>0.05±0.02</td>
<td>-0.06±0.02</td>
</tr>
<tr>
<td>Superior mesenteric arteries</td>
<td>-0.11±0.01</td>
<td>0.10±0.02</td>
<td>-0.08±0.01</td>
<td>-0.09±0.01</td>
<td>0.09±0.01</td>
<td>-0.10±0.02</td>
<td>-0.11±0.01</td>
<td>0.10±0.01</td>
<td>-0.10±0.02</td>
</tr>
<tr>
<td>Large mesenteric arteries</td>
<td>-0.05±0.02</td>
<td>0.10±0.02</td>
<td>-0.07±0.02</td>
<td>-0.05±0.02</td>
<td>0.05±0.02</td>
<td>-0.05±0.02</td>
<td>-0.05±0.02</td>
<td>0.06±0.02</td>
<td>-0.05±0.02</td>
</tr>
<tr>
<td>Adrenal medulla (NE)</td>
<td>-0.07±0.02</td>
<td>0.06±0.02</td>
<td>-0.05±0.01</td>
<td>-0.05±0.01</td>
<td>0.05±0.02</td>
<td>-0.05±0.02</td>
<td>-0.07±0.02</td>
<td>0.05±0.02</td>
<td>-0.05±0.02</td>
</tr>
<tr>
<td>Adrenal medulla (EPI)</td>
<td>-0.04±0.01</td>
<td>0.03±0.01</td>
<td>-0.05±0.01</td>
<td>-0.03±0.01</td>
<td>0.03±0.01</td>
<td>-0.03±0.01</td>
<td>-0.04±0.01</td>
<td>0.03±0.01</td>
<td>-0.03±0.01</td>
</tr>
<tr>
<td>Values are the mean for six rats, ±SEM for k and the 95% confidence limits for turnover rates. DOCA/saline, deoxycorticosterone acetate+saline; NE, norepinephrine; EPI, epinephrine. *p&lt;0.05 vs. untreated rats.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
rats, which developed mild to moderate hypertension, were used. In contrast, previous studies used young rats, which developed severe hypertension. The ability of prazosin to normalize the blood pressure of the DOCA/saline-treated rats in the present study indicates that the hypertension was indeed dependent on sympathetic activity but not necessarily on neuronal hyperactivity. DOCA/saline-treated rats display an increased vascular pressor responsiveness to NE.33,34 It is possible, therefore, that in the present experiments with only mild to moderate hypertension, increased vascular responsiveness to NE was sufficient to produce the level of hypertension observed. In the young rats that developed severe hypertension for 220 versus 160 mm Hg in the present study), increased sympathetic neuronal activity may cause the increased severity of the hypertension.

In the present study, the ability of saline intake to mimic the effect of pressure overload without any increase in pressure raises the possibility that sodium may be involved in the cellular response to pressure load. This is further supported by the finding in renal hypertensive rats that a reduction in sodium intake reverses both the hypertrophy and isomyosin changes33 produced by the increased pressure, without reducing the blood pressure. This indicates that even though sodium restriction did not reduce the

TABLE 5. Plasma Catecholamines in Untreated, Saline-Treated, and Deoxycorticosterone Acetate-Saline-Treated Rats After 3 Weeks of Treatment

<table>
<thead>
<tr>
<th>Group</th>
<th>Norepinephrine (pg/ml)</th>
<th>Epinephrine (pg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Untreated</td>
<td>297±40</td>
<td>166±18</td>
</tr>
<tr>
<td>Saline</td>
<td>222±49</td>
<td>151±12</td>
</tr>
<tr>
<td>DOCA/saline</td>
<td>309±46</td>
<td>243±23*</td>
</tr>
</tbody>
</table>

Values are mean±SEM (n=6 for untreated and saline-treated rats, and n=7 for DOCA/saline-treated rats). DOCA/saline, deoxycorticosterone acetate + saline.

*p<0.05 vs. untreated and saline-treated rats.
increased wall stress in the ventricle, it can decrease the cellular response to the increased wall stress. Moreover, in normotensive rats, sodium restriction caused an increase in V₁ and a decrease in V₂ myosin phenotypes, changes opposite to those produced by increased pressure. How dietary sodium could influence cellular responses involved in cardiac growth is still uncertain. In vitro, stretch-induced sodium influx may be an early signal in the transduction of cardiac load into growth. It is tempting to speculate that increased dietary sodium intake results in increased intracellular myocyte sodium, thus leading to a larger LV mass for a given pressure load. Higher pressures in the LV than the RV may make the LV more sensitive in this regard, thus explaining the more marked response of the LV to increased sodium intake. However, this proposed mechanism is clearly speculative at this point.

In conclusion, the present results demonstrate that saline and DOCA/saline cause concentric LV hypertrophy (i.e., increased wall thickness with no change in internal diameter) such as that produced by pressure overload but without evidence of increased pressure in the case of saline. The absence of increases in filling pressures, cardiac sympathetic neuronal activity, or pressure load during the development of sodium-induced cardiac hypertrophy indicate that in this type of hypertrophy sodium may directly influence cellular mechanisms involved in the regulation of LV mass. If sodium exerts similar effects in humans, increased dietary sodium intake may increase cardiovascular risk both by effects on blood pressure and by direct effects on cardiac mass, considering that LV hypertrophy has emerged as an independent risk factor for cardiovascular morbidity and mortality.

References

18. Kohlmann O Jr, Bresnahan M, Gavras H: Central and peripheral indices of sympathetic activity after blood pressure lowering with enalapril (MK-421) or hydralazine in normotensive rats. Hypertension 1984;6(suppl I):I-1–I-6
35. Sen S, Young DR: Role of sodium in modulation of myocardial hypertrophy in renal hypertensive rats. *Hypertension* 1986;8:918–924

**Key Words** • cardiac hypertrophy • sodium • volume overload • cardiac sympathetic activity
Sodium-induced cardiac hypertrophy. Cardiac sympathetic activity versus volume load.
N G Fields, B X Yuan and F H Leenen

doi: 10.1161/01.RES.68.3.745

Circulation Research is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1991 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7330. Online ISSN: 1524-4571

The online version of this article, along with updated information and services, is located on the
World Wide Web at:
http://circres.ahajournals.org/content/68/3/745

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation Research 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 Circulation Research is online at:
http://circres.ahajournals.org//subscriptions/