Effects of Long-term Air Jet Noise and Dietary Sodium Chloride in Borderline Hypertensive Rats

Diane C. Tucker, Rachel A. Hunt

The hypothesis that simultaneous exposure to a high (8%) sodium chloride diet and behavioral stress (air jet noise) would act synergistically to increase blood pressure was investigated in male borderline hypertensive rats. Rats were fed either a 1% or an 8% sodium chloride diet beginning at 6 weeks of age. Rats in the Air Noise condition were restrained and exposed to random blasts of air jet noise for 2 h/d, 5 d/wk, from 7 to 17 weeks of age. Controls either were placed in identical restrainers and test chambers but not exposed to air jet noise (Restrained Control) or were left undisturbed (Maturation Control). Biweekly indirect blood pressure measurements showed that by 17 weeks of age, the high-sodium chloride diet and air jet noise exposure produced additive increases in blood pressure. Direct blood pressure measurements at 18 weeks of age confirmed the higher systolic pressures in borderline hypertensive rats exposed to both an 8% sodium chloride diet and air jet noise. After ganglionic blockade, the blood pressure of rats in the Air Noise group remained higher than that of Restrained and Maturation Controls, suggesting that the increased blood pressure of air jet noise–exposed rats was not maintained by increased autonomic activity. Blood pressure after maximal vasodilation by hydralazine was increased in rats exposed to both an 8% sodium chloride diet and air jet noise compared with other groups. Baroreceptor reflex sensitivity (tested by graded doses of angiotensin II) did not differ among groups. In summary, exposure to both an 8% sodium chloride diet and air jet noise increased blood pressure in borderline hypertensive rats; however, there was little evidence of synergy, ie, potentiation of the effects of air jet noise by exposure to a high–sodium chloride diet. (Hypertension. 1993;22:527-534.)

KEY WORDS • angiotensin II • sympathetic nervous system • hypertension, genetic • ganglionic blockers

Behavioral stress and high sodium chloride (NaCl) intake each are thought to interact with an individual's genetic background to increase the risk for hypertension in human populations. These relations have been studied experimentally using rat strains with a genetic predisposition to develop hypertension. The superimposition of two environmental risk factors (eg, behavioral stress and high dietary NaCl) on a background of genetic predisposition has been hypothesized to act synergistically to produce hypertension. Synergy may be evidenced by the observation of hypertension only when both risk factors are present or by an increase in the rapidity with which blood pressure rises. High dietary NaCl and behavioral stress may act on common control systems to cause increased blood pressure. The present study used a rodent model with a moderate genetic predisposition to develop hypertension, the borderline hypertensive rat (BHR), to test the hypothesis that simultaneous exposure to long-term behavioral stress (air jet noise) and a high-NaCl diet would produce a synergistic increase in blood pressure. Maintenance of basal blood pressure by autonomic activity and pressor responsiveness to angiotensin II (Ang II) were also assessed.

A sustained increase in blood pressure as a result of long-term behavioral stress or high NaCl intake is rarely observed unless a genetic predisposition to hypertension is present. In the Okamoto spontaneously hypertensive rat (SHR) strain, behavioral stress can accelerate the development of hypertension, but control SHR achieve blood pressures similar to the stressed SHR during the period after the stress is withdrawn. In SHR, the combination of long-term psychosocial stress and a high-NaCl diet appeared to induce a higher blood pressure than either condition alone, although not all conditions were included within the same experiment. Lawler and colleagues and Fisher and Tucker have characterized the F1 cross of SHR female and Wistar-Kyoto male rats (ie, BHR) as a model in which hypertension is reliably produced by behavioral stress or by exposure to a high (8%) NaCl diet and maintained after withdrawal of the stressor. Exposure to intermittent air jet noise was sufficient to increase the blood pressure of weanling (4-week-old) BHR within 2 weeks, indicating sensitivity to long-term behavioral stress during the postweaning period. In contrast, the blood pressures of control BHR usually remain within the high normal range.

Available evidence supports the hypothesis that superimposing behavioral stress on a high-NaCl diet po-
tentiated the pressor response to the behavioral stress.\textsuperscript{4,6,13} In mongrel dogs, Anderson and colleagues\textsuperscript{7,14} demonstrated that long-term avoidance stress in combination with a sodium load (ie, a constant saline infusion) increased blood pressure during 12 days of treatment, although the magnitude of the increase varied substantially among dogs. Neither behavioral stress nor high NaCl alone was sufficient to increase blood pressure in Anderson's studies, providing evidence for a synergy between behavioral stress and high NaCl in producing hypertension. During short-term episodes of exposure to air jet noise (a 30-second air blast to the face), SHR being fed an 8% NaCl diet showed exaggerated blood pressure increases, increased renal sympathetic nerve activity, and increased renal sodium absorption.\textsuperscript{4} Exaggerated pressor responses to cold stress were also observed when SHR were fed an 8% NaCl diet compared with a 1% NaCl diet.\textsuperscript{13} Thus, high levels of dietary NaCl seem to potentiate the cardiovascular and renal responses of SHR to short-term environmental stressors such as air jet noise or cold stress. The present study tested the hypothesis that a similar synergism would be observed during long-term exposure to a high-NaCl diet and behavioral stress in the BHR model.

Methods

Design

At 6 weeks of age, rats were placed individually in hanging cages and assigned to receive either a high (8%) NaCl diet (ICN Biomedicals, Cleveland, Ohio) or to continue to receive the 1% NaCl diet to which they were weaned. At 7 weeks of age, rats assigned to the Air Noise or Restrained Control conditions (see below) began their treatments. Because daily handling and mild restraint may act as stressors, a Maturation Control condition (see below) was also included. Body weights and systolic blood pressures were measured biweekly in rats in the Air Noise and Restrained Control groups beginning at 4 weeks of age until 17 weeks of age. At 18 weeks, direct blood pressure measurements were made in all groups while rats were awake and freely moving in their home cages. During the week before testing, rats were fed a 1% NaCl diet and were not exposed to either air jet noise or restraint. Blood pressure responses to ganglionic blockade and to infused Ang II were measured to provide information about the origin of blood pressure differences among groups.

Animals

Male BHR were used in the present study. BHR were produced in our animal colony by mating salt-sensitive SHR (SHR-S; IBU3 colony\textsuperscript{15}) dams and normotensive Wistar-Kyoto sires purchased from Taconic Farms, Germantown, NY. Within 24 hours of parturition, litters of BHR were culled to a maximum of 10 pups, retaining all male pups. Our animal colony was maintained on a 12-hour light/dark cycle, with lights on at 7 AM. Agway R-M-H 3000 high-protein (30%) diet was fed to SHR-S females throughout gestation and lactation. At 28 days of age, pups were weaned onto Agway R-M-H 1000 diet. Both diets contained 1% NaCl. Throughout the study, food and tap water were available to animals ad libitum. The experimental protocols were approved by the University of Alabama at Birmingham Institutional Animal Care and Use Committee; the animal facilities are fully accredited by the American Association for Accreditation of Laboratory Animal Care.

Air Noise and Restrained Control Conditions

Rats exposed to air noise (Air Noise condition) were placed into 12-in Plexiglas restrainers to restrict movement and then placed into sound-insulated chambers. Random-length pulses of compressed air were used to generate the air noise stress as described previously.\textsuperscript{11} Because rats were placed into restrainers, the principal component of the air noise stress is auditory, not tactile. Duration of both the air pulse and the interpulse interval ranged from 30 to 120 seconds, with the average of each being 75 seconds. The noise level to which BHR were exposed during air jet noise was approximately 120 dB.\textsuperscript{11} Restrained Control rats were placed into identical restrainers and chambers but no air compressor noise was introduced. Stress continued for 2 h/d, 5 d/wk for 10 weeks. Maturation Control rats were housed individually and were not disturbed until direct blood pressure recordings were made at 18 weeks of age.

Indirect Blood Pressure Measurements

Blood pressure and body weight measurements were taken before exposure to air jet noise or restraint. Systolic pressure was measured indirectly from the tail artery using a photoelectric sensor. Signals were amplified with a model 29 amplifier (IITC Inc, Woodland Hills, Calif) and recorded on a model 7D polygraph (Grass Instrument Co, Quincy, Mass). Animals were placed in Plexiglas restrainers but were neither heated nor anesthetized before recording. Systolic pressure was calculated to be the mean of three artifact-free recordings made while the animal was calm. Systolic pressure was determined biweekly from 4 to 16 weeks of age, with a final measurement taken at 17 weeks of age.

Direct Blood Pressure Measurements

After the 17-week body weight and indirect blood pressure measurements, all rats were fed a 1% NaCl diet, and the exposure to air jet noise and restraint was terminated. Within 5 to 7 days, each animal was anesthetized with a mixture of ketamine and xylazine (100 and 7.5 mg/kg IP, respectively), and the femoral artery was cannulated with polyethylene (PE-50) tubing for blood pressure and heart rate determinations. The femoral vein was cannulated with PE-50 tubing for drug administration. Both cannulas were filled with heparinized saline (200 U/mL), plugged, and tunneled subcutaneously to exit between the scapulae. Animals were placed individually in plastic cages (20x25x45 cm) and allowed at least 24 hours to recover from surgery before testing.

On the day of testing, the arterial cannula was connected to a model CP-01 pressure transducer (Century Technology Co Inc, Inglewood, Calif) and flushed with heparinized saline (20 U/mL). Blood pressure and heart rate were recorded on a Grass model 7D polygraph from awake and unrestrained animals in their home cages. Animals were allowed to habituate for approximately 45 minutes before the onset of recording. Samples of blood pressure and heart rate were taken.
every 5 minutes for 20 minutes. Baseline blood pressure and heart rate were the mean of these five samples. Mean arterial pressure (MAP) was calculated using the formula MAP=DBP+1/3(SBP-DBP), where DBP is diastolic blood pressure and SBP is systolic blood pressure. Heart rate was determined with a Grass cardiota-

chometer. Pulse pressures were at least 45 mm Hg for all rats that contributed data. The values measured for systolic and diastolic blood pressures are likely to be dampened relative to the actual pressures. There is, however, no reason to expect differential dampening among treatment groups; therefore, systolic and diastolic blood pressures are reported when they pertain to specific hypotheses.

To test the hypothesis that sensitivity to Ang II is altered by long-term high-NaCl diet, long-term behavioral stress, or their combination, blood pressure and heart rate responses to graded bolus infusions of Ang II (5, 10, 20, 40, and 80 ng/kg IV, Sigma Chemical Co, St Louis, Mo) were measured. The maximum increase in blood pressure and maximum decrease in heart rate were scored. Up to 20 minutes was allowed between injections for blood pressure to return to baseline. Baroreceptor reflex sensitivity was tested by calculating the regression of heart rate decreases on blood pressure increases for each animal.

After blood pressure had returned to baseline following the final dose of Ang II (80 ng/kg IV), chlorisondamine (2.5 mg/kg IV, CIBA-GEIGY, Suffren, NY) was given to determine the contribution of autonomic (largely sympathetic) tone to the maintenance of basal blood pressure in each experimental group. Although blood pressure was allowed to return to baseline before chlorisondamine treatment, any residual changes in autonomic activity after the angiotensin infusions would contribute to this estimate of autonomic tone. Responses to ganglionic blockade with chlorisondamine were measured 20 minutes after injection. This dose and time point were determined in preliminary and previous studies to provide a maximal, stable blockade of blood pressure and heart rate responses to infused phenylephrine in BHR.

To separate the response to Ang II from its facilitation of sympathetic activity, the graded doses of angiotensin were repeated in the context of ganglionic blockade. After the final dose of Ang II (80 ng/kg IV), hydralazine (1.0 mg/kg IV, Sigma) was administered to determine blood pressure in the context of maximal vasodilation. Responses to hydralazine were measured 10 minutes after injection. This dose and time point were determined in preliminary and previous studies to provide a maximal, stable decrease in blood pressure in BHR.

Organ Weights

One to 2 days after blood pressure and heart rate testing, animals were killed by an overdose of anesthe-

sia. The adrenals, kidneys, heart, and retroperitoneal fat pads were removed, blotted, and weighed to the nearest 0.1 mg. Fat and excess tissue were removed from organs before weighing. The left and right adrenals, the two kidneys, and the two fat pads were combined when weighed. The great vessels were trimmed close to the heart, leaving both atria intact. With the exception of the retroperitoneal fat pads, all weights were taken after the organs had been frozen on dry ice. Heart weight was expected to be increased in groups in which blood pressure was chronically elevated. Adrenal weight was expected to be increased by the long-term behavioral stress (air jet noise), and kidney weight was expected to be increased by long-term high (8%) dietary NaCl. The retroperitoneal fat pads were weighed to determine whether behavioral stress or high dietary NaCl altered body composition.

Statistical Analysis

Data are expressed as mean±SEM. No more than two littermates contributed to any experimental group. Indirect blood pressure and body weight data were analyzed by separate profile analyses. Profile analysis is a multivariate analysis that tests whether the pattern of measures (eg, blood pressures or body weights) differs among groups without violating the compound symmetry assumption (ie, that the covariances between all measures are equal). In data collected across time, the covariances between adjacent measures are nearly always more highly correlated than temporally distant measures. Profile analysis also tests whether the overall level differs among groups. Baseline blood pressure was analyzed by a multivariate analysis of variance (MANOVA), with systolic and diastolic blood pressures as the correlated dependent measures. Mean arterial pressure responses to infused Ang II were analyzed by profile analysis, with separate profile analyses conducted on responses before and after administration of the ganglionic blocker. To examine baroreceptor reflex sensitivity, the slope and intercept of the regression of the change in heart rate on the change in blood pressure were calculated for each animal and compared among groups by analysis of variance (ANOVA). Blood pressure after ganglionic blockade was analyzed by MANOVA, with systolic and diastolic blood pressures as the correlated dependent measures. Diastolic blood pressure after hydralazine treatment was compared among groups by ANOVA. Organ weight data were expressed either as absolute weight or as a ratio of organ weight to body weight at time of death (eg, grams per 100 g body weight) and analyzed in separate ANOVAs. Student-Newman-Keuls tests were used to follow up main effects, and simpler analyses were used to pursue interactions.

Results

Indirect Blood Pressure Measurements

The indirect blood pressures of rats later assigned to Air Noise and 8% NaCl groups did not differ from controls at either 4 or 6 weeks of age (P>.50, see Table 1). Consistent with our previous studies, long-term air jet noise exposure increased systolic blood pressure [F(1,41)=8.19, P<.01 for the profile analysis of average pressures observed across time points; see Table 1]. Profile analysis indicated that the effect of air jet noise exposure on the pattern of blood pressure changes did not differ between BHR fed a 1% versus an 8% NaCl diet (P>.20). At 17 weeks of age, BHR exposed to air jet noise and to an 8% NaCl diet had significantly higher systolic blood pressures compared with their respective controls [F(1,41)=6.43, P<.02 for stress and F(1,41)=4.76, P<.04 for diet; see Table 1 and Fig 1].
These data suggest that air jet noise and a high-NaCl diet each increased blood pressure in BHR but do not support a synergism between dietary NaCl and behavioral stress.

Direct Blood Pressure Measurements

Consistent with the observations made using indirect blood pressure measurements, BHR in the Air Noise group had increased baseline mean arterial pressures [F(2,70)=3.53, P<.04; see Table 2], with both systolic and diastolic blood pressures being significantly elevated compared with Restrained and Maturation Controls [F(2,70)=3.87, P<.03 for systolic pressure and F(2,70)=3.21, P<.05 for diastolic pressure]. BHR fed an 8% NaCl diet had increased baseline mean arterial pressures [F(1,70)=4.34, P<.05], with systolic pressure [F(1,70)=6.15, P<.02] but not diastolic pressure [F(1,70)=3.28, P=.07] being significantly elevated compared with BHR fed a 1% NaCl diet. Analysis of baseline systolic, diastolic, and mean arterial blood pressures by MANOVA indicated no significant statistical interaction between behavioral stress and dietary NaCl.

After ganglionic blockade, the mean arterial pressure of rats in the Air Noise group was higher than that of Restrained and Maturation Controls [F(2,59)=5.10, P<.01], with both systolic and diastolic blood pressures being significantly elevated (see Table 2). This suggests that the higher blood pressure observed in rats exposed to air jet noise was not mediated by increased autonomic activity. However, the fall in blood pressure after ganglionic blockade did not differ among the Air Noise and control groups, suggesting that autonomic tone was not changed by long-term exposure to behavioral stress. The fall in mean arterial pressure after ganglionic blockade was smaller in Maturation Controls than in other groups [F(2,59)=3.86, P<.03 for the interaction between behavioral stress and dietary NaCl in mean arterial pressure response to chlorisondamine; see Table 2].

To examine the possibility that structural changes in the vasculature may contribute to the increased blood pressure observed in rats exposed to long-term air jet noise or high dietary NaCl, we measured diastolic blood pressure after maximal vasodilation with hydralazine. There was a significant statistical interaction between behavioral stress and dietary NaCl after hydralazine treatment [F(2,55)=4.28, P<.02; see Table 2]. Follow-up analyses indicated that in BHR fed a 1% NaCl diet, exposure to long-term air jet noise did not affect diastolic blood pressure measured after hydralazine. In contrast, in BHR fed an 8% NaCl diet, rats in the Air Noise group had higher diastolic pressures after hydralazine than Maturation Controls [F(2,22)=3.55, P<.05]. Higher diastolic pressures after hydralazine treatment suggest that structural changes in the vasculature (ie, increased peripheral resistance) contribute to the increased blood pressure in BHR exposed to both air jet noise and a high-NaCl diet.

Profile analysis indicated that neither high dietary NaCl nor exposure to air jet noise affected that pattern of pressor responses to infused Ang II before ganglionic blockade (P>30). BHR fed an 8% NaCl diet, however, had higher mean arterial pressures in response to Ang II compared with BHR fed a 1% NaCl diet [F(1,57)=4.20,

---

**TABLE 1. Systolic Blood Pressures**

<table>
<thead>
<tr>
<th>Age, wk</th>
<th>1% NaCl</th>
<th>Air Noise</th>
<th>8% NaCl</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Restrained Control</td>
<td>Air Noise</td>
<td>Restrained Control</td>
</tr>
<tr>
<td>4</td>
<td>98±4</td>
<td>95±5</td>
<td>93±5</td>
</tr>
<tr>
<td>6</td>
<td>121±3</td>
<td>127±5</td>
<td>118±5</td>
</tr>
<tr>
<td>8</td>
<td>135±6</td>
<td>142±3</td>
<td>140±4</td>
</tr>
<tr>
<td>10</td>
<td>137±5</td>
<td>142±4</td>
<td>137±7</td>
</tr>
<tr>
<td>12</td>
<td>148±6</td>
<td>150±4</td>
<td>139±4</td>
</tr>
<tr>
<td>14</td>
<td>149±6</td>
<td>153±4</td>
<td>147±8</td>
</tr>
<tr>
<td>16</td>
<td>145±3</td>
<td>152±3</td>
<td>149±5</td>
</tr>
<tr>
<td>17*†</td>
<td>142±3</td>
<td>148±3</td>
<td>146±6</td>
</tr>
</tbody>
</table>

Values are mean±SEM, in millimeters of mercury.
*P<.05, Air Noise vs Restrained Control.
†P<.05, 8% NaCl vs 1% NaCl.

---

**Indirect Blood Pressures**

**17 Weeks of Age**

![Graph showing systolic blood pressure (SBP) measured by tail-cuff plethysmography after 10 weeks of exposure to experimental conditions (ie, at 17 weeks of age). Both long-term air jet noise exposure (Air Noise) and 8% NaCl diet significantly increased SBP. Data are mean±SEM. Numbers in parentheses indicate number of rats per group.**

---

Downloaded from http://hyper.ahajournals.org/ by guest on June 4, 2017
Air Noise group showed a greater sensitivity to Ang II compared with Restrained and Maturation Controls.

P<.05; see Fig 2]. BHR in the Air Noise group increased compared with Maturation Controls, and Restricted increased compared with Maturation Controls (P>.20).

When the graded doses of Ang II were repeated after ganglionic blockade to isolate the pressor effects of Ang II from potentiation of sympathetic activity, behavioral stress significantly affected the pattern of pressor responses. Dietary NaCl did not alter the pattern of pressor responses to Ang II after ganglionic blockade (P>.10). As expected, chlorisondamine treatment blocked the bradycardic responses to the Ang II-induced increases in blood pressure. The statistical interaction between dietary NaCl and behavioral stress did not achieve significance, suggesting that an 8% NaCl diet did not potentiate or attenuate the effects of long-term behavioral stress on responses to Ang II.

### TABLE 2. Blood Pressures and Heart Rates at 18 Weeks of Age

<table>
<thead>
<tr>
<th></th>
<th>Maturation Control (n=13)</th>
<th>Restrained Control (n=12)</th>
<th>Air Noise (n=11)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>SBP, mm Hg</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Baseline</td>
<td>151±2</td>
<td>158±2</td>
<td>164±4</td>
</tr>
<tr>
<td></td>
<td>162±6</td>
<td>155±2</td>
<td>173±6</td>
</tr>
<tr>
<td><strong>DBP, mm Hg</strong></td>
<td>99±2</td>
<td>107±2</td>
<td>110±3</td>
</tr>
<tr>
<td></td>
<td>107±5</td>
<td>106±4</td>
<td>117±5</td>
</tr>
<tr>
<td><strong>MAP, mm Hg</strong></td>
<td>118±2</td>
<td>124±2</td>
<td>128±3</td>
</tr>
<tr>
<td></td>
<td>125±5</td>
<td>122±2</td>
<td>135±6</td>
</tr>
<tr>
<td><strong>HR, bpm</strong></td>
<td>333±7</td>
<td>357±11</td>
<td>356±8</td>
</tr>
<tr>
<td></td>
<td>314±8</td>
<td>326±15</td>
<td>328±9</td>
</tr>
</tbody>
</table>

#### Chlorisondamine, 2.5 mg/kg

| SBP, mm Hg           | 105±3                     | 96±5                      | 110±3           |
|                      | 93±4                      | 100±4                     | 120±8           |
| **DBP, mm Hg**       | 70±3                      | 63±3                      | 72±3            |
|                      | 61±2                      | 66±4                      | 78±7            |
| **MAP, mm Hg**       | 82±3                      | 74±4                      | 84±3            |
|                      | 72±3                      | 77±4                      | 92±8            |
| **HR, bpm**          | 305±4                     | 326±6                     | 319±5           |
|                      | 303±9                     | 299±7                     | 306±10          |

#### Hydralazine, 1.0 mg/kg

| SBP, mm Hg           | 84±3                      | 78±4                      | 81±3            |
|                      | 76±3                      | 80±4                      | 92±6            |
| **DBP, mm Hg**       | 53±2                      | 46±3                      | 50±2            |
|                      | 46±3                      | 47±3                      | 57±4            |
| **MAP, mm Hg**       | 64±2                      | 56±3                      | 60±2            |
|                      | 56±3                      | 58±3                      | 68±5            |
| **HR, bpm**          | 353±12                    | 373±13                    | 353±11          |
|                      | 325±16                    | 356±21                    | 335±18          |

SBP indicates systolic blood pressure; DBP, diastolic blood pressure; MAP, mean arterial pressure; HR, heart rate; and bpm, beats per minute. Values are mean±SEM.

P<.05 for the profile analysis of average pressures observed across doses; see Fig 2]. The higher mean arterial pressures observed during Ang II infusion in BHR fed an 8% NaCl diet were due to significantly higher systolic pressures and a borderline significant increase in diastolic pressures (see Table 2). There was no evidence that baroreceptor reflex sensitivity was significantly altered by exposure to air jet noise or by high dietary NaCl (P>.20).

The largest difference among groups in the magnitude of the heart rate change per millimeters of mercury of blood pressure change (ie, the slope) was within the Air Noise condition; air jet noise-exposed rats fed 8% NaCl showed the highest sensitivity (ie, slope: -1.05±0.33), whereas air jet noise-exposed rats fed 1% NaCl showed the lowest sensitivity (slope: -0.54±0.26). The intercepts did not differ significantly from zero (5.37±6.86 and -8.76±6.14, respectively).

When the graded doses of Ang II were repeated after ganglionic blockade to isolate the pressor effects of Ang II from potentiation of sympathetic activity, behavioral stress significantly affected the pattern of pressor responses. Dietary NaCl did not alter the pattern of pressor responses to Ang II after ganglionic blockade (P>.10). As expected, chlorisondamine treatment blocked the bradycardic responses to the Ang II-induced increases in blood pressure. The statistical interaction between dietary NaCl and behavioral stress did not achieve significance, suggesting that an 8% NaCl diet did not potentiate or attenuate the effects of long-term behavioral stress on responses to Ang II.

#### Body Weights

The body weights of rats later assigned to Air Noise and 8% NaCl groups did not differ from controls at 6 weeks of age [143 versus 147 g; F(1,60)=0.57, P>.40]. Although the 8% NaCl diet did not affect the pattern of weight gain, exposure to long-term air jet noise compromised growth [F(6,26)=2.69, P<.04, see Fig 3], especially after prolonged stress (ie, the largest difference in weight gain occurred between 14 and 16 weeks of age). At 18 weeks of age, the body weights of Air Noise and Restrained Control rats were not significantly different from those of Maturation Controls (P>.20). Body weights at 18 weeks of age were lower in BHR being fed 8% NaCl than in BHR fed 1% NaCl [F(1,50)=4.21, P<.05; see Table 3].

#### Organ Weights

Organ weight data are presented in Table 3. Compared with both Restrained and Maturation Controls, exposure to long-term air jet noise increased absolute adrenal weights and the ratio of adrenal weight to body weight [F(2,50)=4.38, P<.02 and F(2,50)=5.82, P<.01, respectively]. High dietary NaCl increased absolute weight of the kidneys and the ratio of kidney weight to body weight [F(1,50)=13.02, P<.001 and F(1,50)=35.27, P<.0001, respectively]. Absolute and relative heart weights were also increased by exposure to high dietary NaCl [F(1,50)=9.16, P<.005 and F(1,50)=14.70, P<.001, respectively]. In addition, the ratio of heart weight to body weight was increased by long-term behavioral stress [F(2,50)=4.10, P<.03], with the Air Noise group increased compared with both Restrained and Maturation Controls, and Restrained increased compared with Maturation Controls.

As observed in our previous study, high dietary NaCl decreased the absolute and relative weights of the retro-
Peritoneal fat pads $F(1,50)=22.42, P<.0001$ and $F(1,50)=22.06, P<.0001$, respectively, suggesting that long-term ingestion of a high-NaCl diet altered body composition in BHR.

Discussion

Although both long-term exposure to a high NaCl diet and behavioral stress increased blood pressure in BHR, the predicted synergy between these environmental contributors to hypertension was not observed. Instead, the blood pressure increases observed with long-term exposure to air jet noise and an 8% NaCl diet were additive and maintained for at least 1 week after the treatments ended. An exception to the additivity of the effects of behavioral stress and dietary NaCl was the increased diastolic pressure after maximal vasodilation only in BHR exposed to both air jet noise and an 8% NaCl diet, suggesting structural changes in the peripheral vasculature in this group. The elevated blood pressure of BHR in the Air Noise group was not dependent on sympathetic activity; ie, compared with the control groups, the blood pressure of air jet noise–exposed BHR remained elevated after ganglionic blockade. Pressor responses to Ang II in the context of ganglionic blockade were larger in groups exposed to air jet noise. Increased pressor responses to infused Ang II could result from a larger number of Ang II receptors, an increase in coupling efficiency, or a change in the receptor subtype distribution.

Although sympathetic tone (estimated by ganglionic blockade) was not increased after long-term exposure to air jet noise or an 8% NaCl diet, increased sympathetic activity may have mediated the initial increase in blood pressure. The hypertensive parent strain of BHR, SHR, shows a sympathetically mediated cardiovascular reactivity, which is most pronounced during the immediate postweaning period. Winternitz and Oparil reported that ingestion of an 8% NaCl diet increased sympathetic activity in 7-week-old SHR. A recent study by Lawler and colleagues demonstrated that the pressor responses of BHR to short-term behavioral stress were exaggerated after a brief (2-week) exposure to a high-NaCl diet; in contrast, BHR fed an 8% NaCl diet for either 2 or 6 months showed decreased reactivity to behavioral stress, even though their baseline blood pressures were elevated. Sympathetic activity is increased before weaning in BHR and could mediate the cardiovascular responses to environmental stressors in young BHR.

When BHR were exposed to air jet noise beginning at 4 weeks of age, blood pressure increased significantly within 2 weeks. In the present study, profile analysis indicated that average systolic blood pressure (measured by tail cuff) observed across time points was increased by long-term exposure to air jet noise that began at 7 weeks of age. ANOVA, however, indicated that the increase in blood pressure was not statistically reliable until 17 weeks of age. This difference between our studies is consistent with cardiovascular reactivity being most pronounced in young (ie, weaning) BHR.

Long-term exposure to a high-NaCl diet has previously been reported to increase blood pressure in BHR. In the present study, both indirect and direct blood pressure measurements confirmed that BHR fed an 8% NaCl diet had higher blood pressures compared with BHR fed a 1% NaCl diet. Although the magnitude of blood pressure increases in response to an 8% NaCl diet reported in the present study is similar to that reported in a recent study from our laboratory, it is lower than that reported by other laboratories. A possible reason for this finding is that BHR in both the Air Noise and Restrained Control groups were exposed to mild restraint for 2 h/d, 5 d/wk for 10 weeks in the present study. Restrained Control rats may have to mild restraint for 2 h/d, 5 d/wk for 10 weeks in the present study. Restrained Control rats may have...)
strait, as did the Air Noise group, but did not experience the unpredictable blasts of air compressor noise. We did not document the hypertension in SHR-S dams and thus cannot be certain that the level of maternal hypertension is equivalent to that in other studies.\textsuperscript{2,23} These and other factors (eg, weaning at 28 days, housing, and husbandry) may contribute to differences in the overall magnitude of blood pressure increases reported among laboratories. It is important to emphasize, however, that although the magnitude of the effect is lower in our study, the direction of the effect is the same as in other studies.

Long-term exposure of BHR to behavioral stress was reported by Lawler and colleagues\textsuperscript{24} to decrease baroreceptor reflex sensitivity. In contrast, exposure of BHR to an 8% NaCl diet from weaning through 17 weeks of age was reported by DiBona and Jones\textsuperscript{25} not to alter baroreceptor reflex control of heart rate or renal sympathetic nerve activity. The present study supported the conclusions of DiBona and Jones\textsuperscript{25} that an 8% NaCl diet did not affect baroreceptor reflex sensitivity in BHR. Although nonsignificant, the trend toward decreased baroreceptor reflex sensitivity in 1% NaCl-fed, air noise–exposed BHR was in the direction predicted by the report of Lawler et al.\textsuperscript{24}

Several lines of evidence suggest that increased renal sympathetic nerve activity may contribute to the development of air jet noise–induced and NaCl-induced hypertension in genetically predisposed individuals. During short-term exposure to air jet noise, SHR show sodium retention mediated by increased renal sympathetic nerve activity.\textsuperscript{4,5,27} Sodium retention during short-term exposure to air jet noise was also reported in BHR rats fed an 8% NaCl diet.\textsuperscript{28} Humans with a genetic predisposition to develop hypertension also showed sodium retention during short-term behavioral stress.\textsuperscript{29}

The extent to which the increased sympathetic nerve activity after short-term behavioral stress would be maintained during repeated stress exposures is not known. Although sympathetic mediation of stress reactivity was not assessed in the present study, tonic sympathetic control of blood pressure was not found to be increased in BHR exposed to long-term air jet noise. Thus, any increases in sympathetic nerve activity that may have occurred during the initial exposure to behavioral stress were not sustained over time. Instead, the long-term elevations in blood pressure were maintained by other factors.

Behavioral stress–induced and dietary NaCl–induced hypertension in BHR can be prevented by disrupting central nervous system control of sympathetic activity. Lesions of the anteroventral third ventricle region, which inhibit both sympathetic efferent activity and vasopressin release,\textsuperscript{30} prevented both stress-induced and NaCl-induced hypertension in BHR.\textsuperscript{31,32} Oparil and colleagues\textsuperscript{2} have demonstrated that SHR-S fail to inhibit sympathetic activity when fed a high-NaCl diet. Reduced anterior hypothalamic area norepinephrine stores and an increased a₂-adrenergic receptor concentration in the anterior hypothalamic area mediate this pattern.\textsuperscript{2} Thus, sympathetic activity (or vasopressin release) may be involved in the development of environmentally induced hypertension in BHR. However,
the present study suggests that after long-term exposure to air jet noise, the increased blood pressure is not maintained by increased sympathetic activity.

Long-term exposure to behavioral stress and a high-NaCl diet were expected to influence responsiveness to Ang II. Induction of hypertension in mice by long-term psychosocial stress results in high levels of circulating renin during the early phase of hypertension, with a return to normal levels during the established phase.33 In the present study, pressor responses to Ang II were assessed after exposure to long-term stress. When interaction between Ang II and sympathetic activity was prevented by ganglionic blockade, increased pressor sensitivity to Ang II was observed only in the Air Noise group. The present study did not test the role of Ang II in maintaining baseline blood pressure or in mediating the stress-induced increases in blood pressure.

In summary, both air jet noise and high dietary NaCl were effective in increasing blood pressure in BHR in the present study. However, these environmental stressors did not act synergistically to potentiate each other. Sympathetic activity did not mediate the environmentally induced increases in blood pressure but may have been important during the development of the increased blood pressure. The data suggest that structural changes in the vasculature are important in maintaining the higher blood pressure of BHR exposed to long-term behavioral stress and high dietary NaCl.

Acknowledgments

This work was supported by R01-HL-39048 from the National Institutes of Health, Bethesda, Md, and the March of Dimes Birth Defects Foundation. D.C.T. is an Established Investigator of the American Heart Association. The authors wish to thank Drs Robert Kirby and Yiu-Fai Chen for their comments on an earlier draft of this manuscript. Jeanette Bicknell provided expert technical assistance. William W. Belsel III provided assistance in data analysis.

References


8. Bicknell provided expert technical assistance. William W. Belsel III provided assistance in data analysis.


Effects of long-term air jet noise and dietary sodium chloride in borderline hypertensive rats.

D C Tucker and R A Hunt

_Hypertension_. 1993;22:527-534
doi: 10.1161/01.HYP.22.4.527

_Hypertension_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 1993 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/22/4/527