Does Vasopressin Contribute to Salt-Induced Hypertension in the Dahl Strain?

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SUMMARY A humoral factor has been implicated in Dahl salt-sensitive genetically hypertensive rats. The goal of this study was to evaluate the pressor role of vasopressin (AVP) in Dahl rats. Salt-sensitive (S) and resistant (R) rats were fed either high (8%) or low (0.4%) NaCl diets for 6 to 8 weeks. Blood pressure was elevated in S rats fed high salt diets (p < 0.05). Plasma AVP increased with high salt diet in both groups (p < 0.05), but was higher in S than R rats (2.0 ± 0.7 and 1.3 ± 0.2 μU/ml respectively, mean ± SE, p < 0.05). With low salt diet, plasma AVP did not differ significantly in S and R rats (1.0 ± 0.2 and 0.7 ± 0.2 μU/ml respectively). Pressor responses to intravenous injection of AVP were greater in S than R rats (p < 0.05), but this difference was also observed with pressor responses to norepinephrine (S > R, p < 0.05); there was no difference in pressor responses to AVP in S rats fed high vs low salt diet. Injection of 50 μg of d(CH₂)₆VDAVP, which selectively inhibits vasoconstrictor effects of AVP, failed to lower blood pressure in S and R rats fed high or low salt diets despite the fact that this dose decreased pressor responses to 8 mU of AVP more than 90%.

Although plasma AVP and vasopressor responses to AVP and NE are slightly elevated in S rats fed high salt, results with d(CH₂)₆VDAVP suggest that vasoconstrictor effects of AVP do not play an important role in the maintenance of hypertension in Dahl S rats. (Hypertension 3: 174-181, 1981)

KEY WORDS • genetic hypertension • d(CH₂)₆VDAVP • vasopressin antagonist • sodium

SELECTIVE inbreeding of rats in the laboratory of Dahl and colleagues resulted in the development of two groups, one that develops hypertension when fed a high sodium diet (sensitive = S) and one that remains normotensive despite a high sodium diet (resistant = R).

Based on studies employing parabiosis, researchers have suggested that a humoral factor(s) is involved in the pathogenesis of hypertension in S rats fed high salt diets. This concept has recently been supported by crossperfusion experiments. The humoral mechanisms could relate to adrenal steroidogenesis, pituitary, or hypothalamic factors.

Several lines of evidence have pointed to arginine vasopressin (AVP) as a possible contributor to salt-induced hypertension in Dahl S rats. First, alterations in adrenal steroidogenesis facilitate 18-hydroxylation of deoxycorticosterone. Thus, levels of 18-hydroxy-deoxycorticosterone are increased in S rats compared to R rats, and this may lead to a net increase in mineralocorticoid activity under the condition of high salt intake. Vasopressin has been implicated in experimentally-induced mineralocorticoid and salt-induced hypertension. Second, the pituitary content of AVP is higher in S rats. Based on these considerations, we advanced the hypothesis that AVP might contribute to hypertension in Dahl S rats on a high salt diet.

This hypothesis was evaluated by measuring 1) plasma levels of AVP; 2) pressor responsiveness to AVP and norepinephrine; and 3) vasodepressor responses to the AVP antagonist, d(CH₂)₆VDAVP.
Methods

The animals for study consisted of 80 female Dahl salt-sensitive (S) and salt-resistant (R) rats from Brookhaven National Laboratory, Upton, New York. The animals were supplied by Dr. Junichi Iwai. All salt-sensitive (S) and salt-resistant (R) rats from these measurements, we calculated an estimate of the consumption of chow and water per day per rat.

Determination of Plasma Levels of Vasopressin

Plasma vasopressin, electrolytes, osmolality, and hematocrit were determined in four groups of animals: S rats, high salt (12 rats); S rats, low salt (13 rats); R rats, high salt (12 rats); and R rats, low salt (14 rats). Blood pressure of these animals was determined by the tail cuff method as described previously.14 Conscious, unperturbed rats were picked up by the tails and immediately decapitated, an approach which minimally affects plasma vasopressin level.14 The blood samples were obtained between 9 and 11 am. Blood was collected from the trunk into heparinized centrifuge tubes placed on ice and capillary hematocrit tubes. Osmolality was measured by freezing point depression. Vasopressin was measured by radioimmunoassay (RIA) as described below.

Sample Preparation

Immediately after collection, heparinized blood was placed on ice and freed of red blood cells by centrifugation at 1400 × g for 15 minutes at 4°C. The plasma was stored at −20°C until prepared for RIA by the technique of Robertson and coworkers.16 Briefly, plasma proteins were precipitated by the addition of 4 ml cold acetone to 1 ml plasma samples, and the precipitate was removed by centrifugation at 3000 × g for 30 minutes at 4°C. The acetone was extracted with 10 ml cold petroleum ether, and phases separated by centrifugation, and the ether phase discarded. The lower aqueous phase containing the AVP was taken to dryness in a 37°C water bath under a stream of room air. The residue was redissolved in 1 ml solution of 0.03% acetic acid, 0.15 M NaCl (Ac-Saline), and stored at −20°C until assay.

Materials

The USP posterior pituitary reference standard was prepared as described in the United States Pharmacopoeia except that chlorobutanol (0.5% by weight) was added as a preservative at the suggestion of Dr. Richard Weitzman of Harbor General Hospital, Torrence, California. This material was stored at 4°C at a concentration of 2.1 units/ml and used as a standard for the RIA. Synthetic AVP obtained from Bachem was also used for secondary standard. Synthetic lysine vasopressin, arginine vasotocin, and oxytocin were obtained from Calbiochem. Purified labile-enzyme-free bovine gamma globulin (BγG) and goat antirabbit gamma globulin were obtained from Miles Research Products. Sephadex G-25 (Pharmacia) was purchased from Sigma.

Antiserum

Rabbit antiserum to AVP obtained by immunization of AVP-thyroglobulin conjugates was prepared locally. The antisera was stored in frozen aliquots of 0.5 ml at −20°C. For each assay, 10 μl of antiserum was diluted 1:100 with 0.5 M Tris buffer. Subsequently this was diluted further, 1:350 with tris buffer and 0.2% BγG, for a final dilution of 1:3,500,000. In the assay tube after addition of other constituents, the effective dilution of antiserum was 1:315,000.

125I- AVP

Iodinated AVP was obtained each month from New England Nuclear, Boston, Massachusetts. NEX-128 vasopressin, 8-arginine, [125I]-monosiodinated has a specific activity of 1200–1500 Ci/μg. The label is attached to tyrosine (H-Cys125I-Tyr-Phe-Gln-Asn-Cys-Pro-Arg-Gly-NH2).

The labeled AVP was stored at 4°C in 0.02 N acetic acid containing 0.1% sodium azide and 0.1% BSA to reduce radiolysis. Under these conditions, the preparation was stable at least 4 weeks.

AVP RIA Utilizing a Nonequilibrium Technique

The 0.2 ml assay buffer (0.05 M Tris-HCl pH 8.0, 0.01 M EDTA, 0.1% sodium azide, 0.2% BγG), 0.2 ml standard or sample (diluted in Ac-Saline), and 0.05 ml AVP antiserum (diluted 1:10,000 in assay buffer) were incubated at 4°C for 36 hours. The 125I- AVP (2000 cpm is 0.05 ml assay buffer) was added, and the incubation continued 24 hours at 4°C. Goat antirabbit gamma globulin (0.1 ml diluted 1:100 in assay buffer containing 1.1% BγG) was added, and the tubes incubated 2 additional hours. The tubes were placed on ice, and the BγG concentration brought to 0.4% by the addition of 0.1 ml assay buffer containing 1.1% BγG. The 125I- AVP bound to antibody was precipitated by the addition of 0.7 ml of 25% polyethylene glycol 6000 and separated from free 125I- AVP by centrifugation at 3000 × g at 4°C for 1 hour. The supernatant is decanted and discarded. Each pellet was counted for 5 minutes in a Micromedic MS 588 gamma spectrometer with a counting efficiency of 76%. Nonspecifically-bound 125I- I- AVP was determined in separate tubes incubated without antiserum.

Standards were assayed in triplicate and unknown samples assayed in duplicate at three different dilutions to assess parallelism. The computer program of Rodbard16 was used to obtain the weighed regres-
Also, the potency of unknowns and the assessment of superimposable inhibition curves: 2.4 pg synthetic 176 HYPERTENSION VOL 3, No 2, MARCH-APRIL 1981 USP-AVP obtained from Dr. Richard Weitzman's laboratory also gave a standard curve superimposable on our USP-AVP curve. Cross reactivities for the related peptides (calculated at 50% B/Bo) were: LVP 27%, AVT 5%, and oxytocin < 0.1%. Reproducibility of the assay was determined by examination of 13 standard curves generated in a 6-month period. These assays utilized six different preparations of 125I-AVP. The ED50 for the standard curves was 2.0 ± 0.27 μU (± SD). The lowest level of AVP producing a statistically significant inhibition (2 SD) of 125I-AVP binding was 0.24 μU/tube. The intraassay coefficients of variation determined in one experiment measuring 36 samples at three volumes averaged 6.5%. Also, it was determined that no detectable inhibition of 125I-AVP binding above background was produced by plasma from Brattleboro rats for the related peptides (calculated at 50% B/Bo).

Recovery of unlabeled AVP (15 to 60 μU) added to 2 ml samples from human plasma pool averaged 82% and showed a highly significant correlation coefficient (r = 0.992, p = 0.001) (n = 14). Recovery of unlabeled AVP (0 to 150 μU) added to pooled rat plasma averaged 72% and showed a highly significant correlation coefficient (r = 0.992, p < 0.01) (n = 21). Recovery of labeled vasopressin in rat plasma averaged 73 ± 2% (± SD) (n = 11). Tabulated values have been corrected for this loss.

The relationship of immunoassayable and bioassayable vasopressin in six samples of rat pituitary was also highly significant (r = 0.773, p < 0.01).

Responses to Vasopressin and d(CH2)6VDAVP in Conscious Rats

These experiments were performed in separate groups of animals consisting of eight S rats fed high salt, seven S rats on low salt, seven R rats on high salt, and seven R rats on low salt. With pentobarbital (35 mg/kg, i.p.) anesthesia, the left femoral artery and vein were catheterized using polyethylene tubing (outer diameter 0.97 mm), the tip of which was tapered to an outer diameter of 0.6 to 0.7 mm for insertion into vessels. The catheters were tunneled subcutaneously to the scruff of the neck and exited through a small incision. Catheters were filled with heparinized saline, plugged with obturators, and passed through larger tubing (outer diameter 3.5 mm) which was fixed to the skin with a wound clip and cranioplastic cement. Following surgery, animals were treated prophylactically with 30,000 units of penicillin intramuscularly. Rats were allowed to recover for 2 days, and regain normal eating and drinking behavior, activity, and body weights.

Before the experiment, animals were placed in a round container 24 cm in diameter and 25 cm high. Blood pressure was recorded from the femoral arterial cannula using a P23Db Statham pressure transducer. Heart rate was monitored using a cardiotachometer triggered by arterial pressure pulsations. These parameters were continuously recorded on a Beckman Dynograph recorder. After 30 to 60 minutes, which allowed blood pressure and heart rate to stabilize at basal levels, the following interventions were performed.

The synthetic analog of vasopressin, d(CH2)6 VDAVP, which specifically blocks the pressor action of vasopressin,17 was injected intravenously in a bolus of 50 μg of (0.2 ml). (The d(CH2)6VDAVP was supplied by Dr. Maurice Manning, Medical College of Ohio, Toledo, Ohio.) After 5 minutes, 8 μL of arginine vasopressin (AVP, Bachem) was injected intravenously to assess the efficacy of blockade of vasopressin. In other experiments, we also confirmed that 5 minutes was sufficient to achieve complete antagonism of the pressor effect of vasopressin after injection of 50 μg of d(CH2)6VDAVP (see results below). Responses to the vehicle alone were tested in a corresponding manner. Glyceryl trinitrate (GTN, 10 μg, 0.2 ml) also was injected intravenously to assess responsiveness of the animals to a vasodepressor stimulus. At the end of experiments, both catheters were refilled with heparinized saline and plugged with an obturator.

After 1 day of recovery during which body weight and baseline hemodynamic state returned to those of the previous day, pressor and heart rate responses to constrictor stimuli were tested. Norepinephrine bitartrate (0.01, 0.05, and 0.1 μg, calculated as the base) and AVP (2 and 8 μL/U) were injected intravenously in a bolus of 0.2 ml. Norepinephrine was administered before vasopressin, and doses were given in graded sequence. Five minutes were allowed between doses of norepinephrine and 20 to 30 minutes allowed between doses of vasopressin so that baseline blood pressure could return to normal levels. Injections of the vehicle of each drug had no detectable effect on blood pressure and heart rate.

In preliminary experiments of five animals (2 S-high salt, 2 S-low salt, 1 R-low salt) we also assessed the efficacy of vasopressin blockade in another manner. In these experiments, we evaluated the magnitude and time course of the pressor response to intravenous AVP, 8 μL/U of which was repeatedly injected at 1- to 4-hour intervals. At the peak of the pressor response to the second injection of AVP, we injected 50 μg of d(CH2)6VDAVP intravenously.

Statistics

Statistical comparisons were performed using one-way analysis of variance followed by Duncan's multiple range test. Values of p < 0.05 were regarded as statistically significant. Pressor responses in S and R rats were compared using analysis of variance and a parallel line bioassay.18

Tabulated values have been corrected for this loss.
**Results**

**Plasma Levels of Vasopressin and Other Baseline Data**

Blood pressure was elevated in S rats fed high salt diet compared to the other groups \((p < 0.05)\) (table 1).

There was no difference in body weight in the four groups of animals.

With low salt diets, plasma AVP did not differ significantly in S and R rats. Plasma AVP increased in both groups with high salt diets \((p < 0.05)\), but was greater in S than R rats \((p < 0.05)\).

Plasma osmolality, sodium, and chloride levels were higher in S rats on high salt compared to S rats fed low salt \((p < 0.05)\). There was no significant difference in these variables in R rats fed high vs low salt diets.

Hematocrit was slightly higher in S rats \((p < 0.05)\), but was not significantly altered by diet in either group.

Consumption of chow and water of S and R rats was greater on high salt compared to low salt diet \((p < 0.05)\). On the high salt diet, S rats consumed 18 ± 2 g of chow (mean ± se) and drank 62 ± 2 ml of water per day, and R rats consumed 22 ± 1 g and drank 61 ± 3 ml per day respectively. On the low salt diet, S rats consumed 15 ± 1 g and drank 29 ± 1 ml of water, and R rats consumed 15 ± 1 g and drank 25 ± 2 ml per day respectively.

**Depressor Response to Vasopressin Analog in Conscious Animals**

Blood pressure was elevated in S rats fed high salt compared to the other groups \((p < 0.05)\) (table 2).

There was no difference in body weight and the heart rates in the four groups of animals.

The vasodepressor responses to 50 \(\mu g\) of \(d(CH_2)_5VDAVP\) are summarized in figure 1. There was no significant reduction in mean arterial blood pressure in any group of animals. However, the pressor responses to 8 \(mU\) AVP was reduced more than 90% by this dose of \(d(CH_2)_8VDAVP\) when compared to the pressor response to 8 \(mU\) AVP injected into the same animals on the next day. Glyceryltrinitrate produced a depressor response in each group.

In the preliminary experiments, efficacy of vasopressin blockade was also confirmed in another manner (fig. 2). After the peak pressor response to the first injection of 8 \(mU\) AVP, which averaged 53 ± 7 mm Hg, blood pressure took 12.8 ± 1.4 minutes before it returned to baseline levels. At the peak pressor response to the second injection of 8 \(mU\) AVP, which averaged 52 ± 6 mm Hg, 50 \(ng\) of \(d(CH_2)_8VDAVP\) was injected intravenously; it normalized the blood pressure within 2.1 ± 0.3 minutes. After 4 to 8 hours from the injection of

**Table 1. Plasma Levels of Vasopressin and Other Baseline Data**

<table>
<thead>
<tr>
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</thead>
<tbody>
<tr>
<td>Body weight (g)</td>
<td>250.6 ± 3.3</td>
<td>255.5 ± 3.8</td>
<td>248.1 ± 4.4</td>
<td>249.1 ± 3.7</td>
<td>ns</td>
</tr>
<tr>
<td>Systolic BP (mm Hg)**</td>
<td>152.2 ± 4.3</td>
<td>118.7 ± 2.6</td>
<td>105.9 ± 2.1</td>
<td>103.5 ± 1.8</td>
<td>SH &gt; SL, RH, RL*; SL &gt; RH, RL*</td>
</tr>
<tr>
<td>Plasma vasopressin ((\mu U/ml))</td>
<td>2.0 ± 0.3</td>
<td>1.0 ± 0.2</td>
<td>1.3 ± 0.2</td>
<td>0.7 ± 0.2</td>
<td>SH &gt; SL, RH, RL*; RH &gt; RL*</td>
</tr>
<tr>
<td>Plasma osmolality (mOsm/liter)</td>
<td>286.8 ± 1.0</td>
<td>282.3 ± 1.2</td>
<td>287.0 ± 2.4</td>
<td>286.4 ± 1.4</td>
<td>SH &gt; SL*</td>
</tr>
<tr>
<td>Plasma Na (mEq/liter)</td>
<td>140.0 ± 0.4</td>
<td>136.3 ± 0.7</td>
<td>134.2 ± 1.3</td>
<td>137.9 ± 1.1</td>
<td>SH, RH &gt; SL*</td>
</tr>
<tr>
<td>Plasma Cl (mEq/liter)</td>
<td>104.9 ± 0.7</td>
<td>101.1 ± 0.7</td>
<td>104.2 ± 1.0</td>
<td>104.1 ± 1.0</td>
<td>SH, RH, RL &gt; SL*</td>
</tr>
<tr>
<td>Hematocrit (%)</td>
<td>42.3 ± 0.4</td>
<td>41.6 ± 0.2</td>
<td>39.8 ± 0.3</td>
<td>39.8 ± 0.2</td>
<td>SH &gt; RH, RL*; SL &gt; RH, RL*</td>
</tr>
</tbody>
</table>

*p < 0.05.

**Table 2. Baseline Data for Conscious Study**

<table>
<thead>
<tr>
<th>Baseline measurement</th>
<th>S-H (8 rats)</th>
<th>S-L (7 rats)</th>
<th>R-H (7 rats)</th>
<th>R-L (7 rats)</th>
<th>Statistical significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body weight (g)</td>
<td>248 ± 3.7</td>
<td>258 ± 7.8</td>
<td>250.8 ± 5.8</td>
<td>246.3 ± 8.5</td>
<td>ns</td>
</tr>
<tr>
<td>Systolic BP (mm Hg)**</td>
<td>165.0 ± 4.5</td>
<td>141.7 ± 7.4</td>
<td>133.3 ± 2.4</td>
<td>136.3 ± 2.3</td>
<td>SH &gt; SL, RH, RL*</td>
</tr>
<tr>
<td>Diastolic BP (mm Hg)**</td>
<td>100.5 ± 2.4</td>
<td>88.6 ± 4.7</td>
<td>81.4 ± 2.5</td>
<td>85.7 ± 1.0</td>
<td>SH &gt; SL, RH, RL*</td>
</tr>
<tr>
<td>Mean BP (mm Hg)**</td>
<td>128.8 ± 3.4</td>
<td>112.0 ± 5.1</td>
<td>104.8 ± 2.8</td>
<td>108.3 ± 1.1</td>
<td>SH &gt; SL, RH, RL*</td>
</tr>
<tr>
<td>Heart rate (beats/min)</td>
<td>389.4 ± 5.2</td>
<td>410.0 ± 10.7</td>
<td>382.9 ± 13.9</td>
<td>416.4 ± 14.6</td>
<td>ns</td>
</tr>
</tbody>
</table>

*p < 0.05.

**Blood pressure was measured by direct intraarterial cannulation.**

S = salt sensitive; R = salt resistant; H = high salt; L = low salt. ns = no significant differences among groups.
d(CH$_2$)$_8$VDAVP, a third injection of 8 mU AVP produced a pressor response similar in magnitude (58 ± 7 mm Hg) and duration (13.3 ± 1.7 min) to the first injection of AVP without AVP antagonist (fig. 2).

**Pressor Response to Arginine Vasopressin and Norepinephrine in Conscious Animals**

Pressor responses to AVP and NE were augmented in S compared to R rats on both high and low salt diets (p < 0.05) (table 3 and fig. 3). Pressor responses to AVP were not altered by low vs high salt diets. In S rats, pressor responses to norepinephrine were slightly augmented by high salt diet. Although pressor responses to AVP and norepinephrine were greater in S rats, heart rate change in response to the injections of norepinephrine and AVP were similar in the four groups of animals.

**Relationship of Pulse Interval and Mean Arterial Pressure in Dahl Rats on Low and High Salt Diets**

The regression (slope) relating pulse interval and mean arterial pressure after norepinephrine injection

![Figure 1](http://hyper.ahajournals.org/)

**Figure 1.** Left Panel Depressor responses to d(CH$_2$)$_8$VDAVP and glyceryl trinitrate (GTN) None of the groups showed significant depressor responses to intravenous d(CH$_2$)$_8$VDAVP (50 µg) Right Panel: Efficacy of vasopressin (AVP) antagonist. This dose of d(CH$_2$)$_8$VDAVP blocked the pressor response of 8 mU AVP by more than 90%. S = salt sensitive, R = salt resistant, H = high salt, L = low salt.

**Figure 2.** Efficacy of vasopressin antagonist. Doses of 8 mU of arginine vasopressin (AVP) were repeatedly injected intravenously. At the peak pressor response to the second injection, 50 µg of d(CH$_2$)$_8$VDAVP was injected intravenously. After 15 minutes, it reduced the blood pressure to baseline. Four hours after the injection of d(CH$_2$)$_8$VDAVP, pressor response to 8 mU AVP became normal.

![Table 3](http://hyper.ahajournals.org/)

**Table 3.** Relative Potency (RP) Values of Pressor Responses to Arginine Vasopressin (AVP) and Norepinephrine (NE) Between Groups

<table>
<thead>
<tr>
<th>Rat groups</th>
<th>RP</th>
<th>95% Confidence limits</th>
<th>RP</th>
<th>95% Confidence limits</th>
</tr>
</thead>
<tbody>
<tr>
<td>S-L vs S-H</td>
<td>1.28</td>
<td>2.47-0.7</td>
<td>1.45*</td>
<td>2.13-1.07</td>
</tr>
<tr>
<td>R-H vs S-H</td>
<td>3.22*</td>
<td>7.58-1.73</td>
<td>3.94*</td>
<td>12.22-2.29</td>
</tr>
<tr>
<td>R-L vs S-L</td>
<td>2.69*</td>
<td>6.77-1.30</td>
<td>2.38*</td>
<td>3.65-1.77</td>
</tr>
<tr>
<td>R-L vs R-H</td>
<td>1.27</td>
<td>1.81-0.61</td>
<td>1.13</td>
<td>1.68-0.78</td>
</tr>
</tbody>
</table>

*Indicates significant differences in pressor responses to agonists (p < 0.05).

S = salt sensitive; R = salt resistant; H = high salt, L = low salt. RP is the ratio of the dose of agonist required to produce a certain response in one group vs the dose required in the second group to produce the same response. For example, 2.2 times more AVP was required in the RH than SH group to produce the same increase in arterial blood pressure. The fact that the 95% confidence limits of the RP do not include the ratio one indicates that AVP produces a significantly greater pressor response in the SH group than in the RH group (see ref. 18).
calculated from the data in figure 3 was significantly less in S than in R rats regardless of diet (fig. 4). Thus, reflex bradycardia during pressor responses to norepinephrine were impaired in S rats compared to R rats.

Discussion

The major findings in this study were: 1) high salt diet increases plasma AVP in Dahl S and R rats with higher levels in hypertensive rats; 2) pressor responses to AVP are greater in S than in R rats, but pressor responses are not different in S rats fed low vs high salt diet; and 3) antagonism of the vasoconstrictor and pressor action of AVP does not decrease blood pressure in Dahl S and R rats.

This discussion will deal with several issues: first, the mechanism of increased AVP; second, the mechanism of augmented pressor responses to AVP; and third, interpretation of the experiments employing the AVP antagonist, d(CH₉)₇VDAVP.

Increased Plasma AVP

The increase in plasma AVP was probably related mainly to high salt diet since there was no difference in AVP in S and R rats fed low salt diet. Moreover, plasma AVP increased with high salt diet in both S and R rats. This is consistent with the observation of Crofton et al.¹ that normal (Long-Evans) rats increased urinary excretion of AVP when they had 1% NaCl for drinking water. However, genetic factors may have had some influence on plasma AVP since AVP was higher in S rats than in R rats when both were fed high salt diet. Rapp and Dahl⁸ have previously reported that the pituitary content of vasopressin is higher in S than in R rats on both high and low salt diet. The mechanism for higher plasma AVP in S rats vs R rats on high salt diet in our study is not clear. Plasma osmolality increased with high salt diet in S rats, but did not increase with high salt in R rats. However, plasma osmolality did not differ significantly in S and R rats on high salt diet. Thus, we cannot conclude that the higher AVP in S rats fed high salt diet resulted from higher plasma osmolality. Impaired baroreflex function in S rats (fig. 4) also may have contributed to elevated plasma AVP,¹⁹ but this was not a major factor since S rats on low salt had impaired baroreflex function and less elevation of AVP than S rats on high salt intake.

Increased Pressor Responses

Pressor responses to AVP were augmented in S rats. This augmentation was observed on low as well as high salt diet. Indeed, there was no difference in pressor responses to AVP in S rats fed high vs low salt diet. Moreover, the augmented pressor responsiveness was not specific for AVP since S rats also exhibited augmented responsiveness to norepinephrine. Dahl et al.²⁰ earlier reported augmented pressor responses to norepinephrine and angiotensin II in S rats, but failed to observe increased pressor responses to AVP in ether-anesthetized rats.
Differences in baseline blood pressure among the groups may not explain the potentiation of the pressor responses to norepinephrine and vasopressin in Dahl S rats. Higher baseline resistance (and blood pressure) would be expected to limit constrictor (and pressor) responses and not potentiate the responses to the constrictor stimuli observed here.18, 21. 22

There are at least two possible explanations for augmented pressor responsiveness to AVP and norepinephrine in S rats. One explanation might be augmented vascular reactivity. Ze cannot exclude this possibility for AVP since we do not have data on vascular responses to local administration of AVP in Dahl rats. However, previous studies in our laboratories have failed to demonstrate augmented vascular reactivity to local administration of norepinephrine and angiotensin II in hindquarters and renal circulation in prehypertensive or early hypertensive Dahl rats.18, 23 Thus, the augmented pressor responsiveness to norepinephrine may not be explained by augmented vascular reactivity.

A second possible explanation for the augmented pressor responsiveness to intravenous injections of AVP and norepinephrine might be impairment of baroreflex buffering. In S rats fed either low or high salt diet, a given increment in arterial pressure was associated with less reflex bradycardia than was observed in R rats (figs. 3 and 4). This suggests impairment of baroreflex control in Dahl S rats. Impairments of baroreflex buffering could contribute to an augmented pressor responsiveness to AVP and norepinephrine without an enhancement of vascular reactivity.

Vasopressin Antagonism

The two observations described above (increased plasma levels of AVP and augmented pressor responses to AVP in S rats) suggested that vasoconstrictor and pressor actions of AVP might contribute to hypertension in the Dahl S rats. To evaluate this possibility, vasopressor responses to an antagonist of vasopressin (d(CH2)8VDAVP) were measured. Contrary to our expectation, the results did not support a pressor role for AVP in hypertensive S rats fed high salt. Vasodepressor responses to d(CH2)8VDAVP (50 μg) were not detected in S or R rats fed low or high salt diet.

Before concluding that these data exclude a significant pressor role for AVP, we should consider the efficacy of the antagonist and the responsiveness of the preparation.

The effectiveness of the antagonist in inhibiting pressor responses to AVP was demonstrated in two ways. The dose was proven to antagonize almost completely the pressor responses to AVP. Furthermore, the antagonist blocked the pressor effects of vasopressin both when given before vasopressin (fig. 1) and when given during late peak of the pressor response to vasopressin (fig. 2). Injection of the vehicle alone had no detectable effect. These results establish that the dose of d(CH2)8VDAVP was adequate and effective in inhibiting the vasopressor influence of vasopressin.

Animals studied in the conscious state also were known to be quite responsive to a second vasodepressor stimulus; injection of glyceryl trinitrate (GTN) produced significant decreases in arterial pressure. These decreases were at least as great in hypertensive S rats fed high salt diet as in the other groups (fig. 1).

Thus, the antagonist of AVP (d(CH2)8VDAVP) was effective and the animals were responsive to a vasodepressor stimulus, but d(CH2)8VDAVP failed to produce a detectable fall in arterial pressure. These findings indicate that the constrictor effects of AVP do not play an important role in the maintenance of salt-induced hypertension in Dahl S rats.

The results of this study do not exclude the possibility that vasopressin may play an important role in the development as opposed to the maintenance of hypertension in Dahl S rats. In another model of salt-induced hypertension, desoxycorticosterone (DOC)-salt hypertension, the failure of production of hypertension in diabetes insipidus rats which genetically have no vasopressin suggests that vasopressin could be necessary for the development of hypertension.12 Moreover, the results of this study do not exclude a role of the renal effects of vasopressin in either the development or maintenance of hypertension in Dahl S rats since d(CH2)8VDAVP may not block the renal (antidiuretic) effects of vasopressin. However, the results do seem to exclude a role of the constrictor effects of vasopressin in the maintenance of hypertension in Dahl S rats.

In summary, the constrictor effects of vasopressin do not contribute to the sustained hypertensive state in Dahl S rats fed a high salt diet. This conclusion is based on the fact that, despite significant elevations in plasma vasopressin and pressor responsiveness to vasopressin, hypertensive S rats fail to achieve a detectable vasodepressor response to adequate blocking doses of the specific vasopressin antagonist, d(CH2)8VDAVP.

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