Huang, Bing S., Hao Wang, and Frans H. H. Leenen. Chronic central infusion of aldosterone leads to sympathetic hyperreactivity and hypertension in Dahl S but not Dahl R rats. Am J Physiol Heart Circ Physiol 288: H517–H524, 2005. First published September 30, 2004; doi:10.1152/ajpheart.00651.2004.—Six-week-old Dahl salt-sensitive (S) and -resistant (R) rats received for 2 wk an intracerebroventricular infusion of aldosterone (Aldo) (22.5 ng/h) or vehicle containing artificial cerebrospinal fluid (aCSF) with 0.15 M Na+. At 8 wk, mean arterial pressure (MAP), heart rate (HR), and renal sympathetic nerve activity (RSNA) were recorded in conscious rats at rest, in response to air stress, and to an intracerebroventricular injection of the α2-adrenoceptor agonist guanabenz or ouabain. Baroreflex control of RSNA and HR was estimated by using intravenous phentolamine and nitroprusside. In Dahl S but not Dahl R rats, Aldo raised MAP by 20–25 mmHg, doubled sympathoexcitatory and pressor responses to air stress and sympathoinhibitory and depressor responses to guanabenz, and impaired baroreflex function. In Dahl S but not Dahl R rats, Aldo significantly increased content of ouabain-like compounds (OLCs) in the hypothalamus and attenuated excitatory responses to ouabain. Aldo did not affect water intake, plasma electrolytes, or OLC in plasma and adrenal glands. In another set of three groups of Dahl S rats, Aldo dissolved in aCSF containing 0.16, 0.15, or 0.14 M Na+ was infused intracerebroventricularly for 2 wk. CSF Na+ concentration ([Na+]c) showed only a nonsignificant increase at resting MAP increased from 111 ± 3 mmHg in rats with Aldo in 0.14 M Na+ to 131 ± 3 and 147 ± 3 mmHg with Aldo in 0.15 and 0.16 M Na+, respectively (P < 0.05 for both). These findings indicate that in Dahl S rats, intracerebroventricular infusion of Aldo causes similar central responses as high salt intake, i.e., increases in brain OLC content, sympathetic hyperreactivity, and hypertension. The extent of the increase in blood pressure (BP) by intracerebroventricular Aldo depends on the [Na+]c in the vehicle. In Dahl R rats, intracerebroventricular Aldo did not increase brain OLC, sympathetic reactivity, and BP, suggesting that in this rat strain, a decrease in central responsiveness to mineralocorticoids may contribute to its salt-resistant nature.

IN DAHL SALT-SENSITIVE (S) rats, high salt intake increases brain ouabain-like compounds (OLCs) (18, 37) followed by activation of the brain renin-angiotensin system (RAS) (20), sympathetic hyperactivity, and hypertension. Stimulation of mineralocorticoid receptors (MR) in the brain contributes to the salt-sensitive hypertension in Dahl S rats, because intracerebroventricular infusion of an MR antagonist prevents hypertension (10). Intracerebroventricular infusion of amiloride or its analog benzamil, which inhibits Na+ channels, increased the brain content of OLC as well as the resting BP in Dahl S rats (37) as well as the hypertension induced by intracerebroventricular infusion of aldosterone in normotensive rats (11). These findings indicate that stimulation of MR in the brain causes hypertension through activation of benzamil-blockable Na+ channels. Amiloride-sensitive Na+ channels in the brain include Phe-Met-Arg-Phe-NH2-gated Na+ channels (18) as well as epithelial Na+ channels (ENaCs) (5, 35).

In the kidney, aldosterone enhances Na+ entry into the collecting duct cells by activation or translocation of preexisting ENaCs and more chronically by induction of these channels (7). ENaCs have also been found in endothelial cells of brain microvessels (35). MRs also exist in the brain, including the choroid plexus and ventricular ependyma (Refs. 33 and 39). Depending on the location of ENaCs in the brain and sites of action of aldosterone, stimulation of ENaCs may increase cerebrospinal fluid (CSF) Na+ concentration ([Na+]c) as well as increase neuronal responsiveness to increases in CSF [Na+]c. Indeed, we recently showed (36) that in Wistar rats, a short-term intracerebroventricular infusion of artificial CSF (aCSF) containing 0.16 M Na+ had no demonstrable effects, but after intracerebroventricular infusion of aldosterone for 2 h, the same intracerebroventricular infusion significantly increased resting renal sympathetic nerve activity (RSNA), blood pressure (BP), and heart rate (HR). These responses were all blocked by intracerebroventricular pretreatment with either benzamil or antibody Fab fragments binding OLC with high affinity (36). A 2-wk intracerebroventricular infusion of aldosterone in aCSF containing 0.15 M Na+ at 25 ng/h significantly increased the brain content of OLC as well as the resting BP in Wistar rats (36). Intracerebroventricular pretreatment with benzamil also abolished these effects (36). These findings suggest that an aldosterone-induced increase in benzamil-blockable Na+ channels in the brain enhances Na+ entry, e.g., from the CSF into the brain interstitial fluid, leading to an increase in brain OLC and, thereby, hypertension. In Dahl S but not Dahl salt-resistant (R) rats, high salt intake causes a persistent increase in CSF [Na+]c (19). In addition, Dahl S rats exhibit enhanced sympathoexcitatory and pressor responses to increased CSF [Na+]c compared with Dahl R rats (21). Considering the above, we hypothesized that mineralocorticoids, possibly produced locally in the brain (13), contribute to enhanced responses to CSF Na+ in Dahl S rats and thereby to sympathoexcitation and hypertension in Dahl S rats on high salt intake. Besides higher mineralocorticoid production and release, increased responses to MR stimulation in Dahl S versus Dahl R rats may play a role. If the latter mechanism is involved, long-term intracerebroventricular administration of aldosterone will mimic the effects of high salt intake and
increase, in parallel, the brain content of OLC, sympathetic activity, and resting BP in Dahl S but not (or to a less extent) in Dahl R rats, and additional Na\(^+\) in the CSF will enhance the responses to intracerebroventricular aldosterone.

To test this concept, we investigated whether 1) in Dahl S rats, a 2-wk intracerebroventricular infusion of aldosterone in aCSF containing 0.15 M Na\(^+\) (just above 0.146 M in Dahl S rats on regular salt, Ref. 19) causes not only hypertension but also an increase in brain OLC, sympathetic hyperreactivity, and impairment of arterial baroreflex function, similar to the responses of this rat strain to high salt intake (1); 2) the extent of the responses to intracerebroventricular aldosterone is related to the [Na\(^+\)] in the aCSF vehicle; and 3) similar to their resistance to dietary salt, Dahl R rats are resistant to intracerebroventricular aldosterone in terms of increases in brain OLC, sympathetic activity, and BP.

### MATERIALS AND METHODS

Male Dahl S and Dahl R rats, 5–6 wk of age, were obtained from Harlan Sprague Dawley (Indianapolis, IN) and housed at constant room temperature, humidity, and light cycle (12:12-h light-dark). Throughout, the animals received regular rat chow (101 \(\mu\)mol Na\(^+\)/g; Harlan Sprague Dawley) and water ad libitum. All experiments were carried out in accordance with the guidelines of the University of Ottawa Animal Care Committee for the care and use of laboratory animals.

#### Protocol 1

After 3–5 days of adaptation, under halothane inhalation, a 23-gauge stainless steel right-angled cannula was implanted into the left lateral cerebroventricle, as previously described (20). The cannula was connected to an osmotic minipump (model 2ML2, lot 10055-02; ALZET) for a 2-wk intracerebroventricular infusion of aldosterone at 22.5 ng/h of vehicle (aCSF with 0.2% ethanol and 0.15 M Na\(^+\)). The pump rate was 4.5 ± 0.1 \(\mu\)l/h ± SD. This dose of aldosterone was based on previous studies (31, 36) in which in Wistar and Sprague Dawley rats intracerebroventricular aldosterone increased BP, sympathetic activity, and brain OLC but had no demonstrable effects on plasma electrolytes, renin, vasopressin, or aldosterone. This rate of infusion is ineffective when given systemically (8, 31). Previous studies (2, 36) using up to 0.5% ethanol in aCSF as vehicle reported no effects on growth or neurological behavior. Infusion of aCSF containing 0.15 M Na\(^+\) at ~4.5 \(\mu\)l/h adds a small amount of Na\(^+\) to the CSF, considering that the CSF [Na\(^+\)] is ~0.146 M in Dahl S (19) and Wistar (20) rats on regular salt intake, and the secretion rate of CSF is ~150–350 \(\mu\)l/h in rats (3). During the same surgery, a 23-gauge guide needle was fixed on the skull of a rat over the right lateral cerebroventricle for intracerebroventricular injections (20). After surgery, rats were provided for 2 wk with tap water and regular rat chow. Body weight and food and water intake were recorded daily.

At the end of the 2-wk intracerebroventricular infusion of aldosterone or vehicle (\(n = 6–8\)/group), with rats under halothane anesthesia, catheters [polyethylene (PE)-10 fused to PE-50] were placed into the left carotid artery. The next morning, in conscious free-moving rats, the carotid arterial catheters were connected to pressure transducers.

#### Protocol 2

Groups of Dahl S and Dahl R rats underwent intracerebroventricular cannulation and then were treated with either aldosterone or vehicle intracerebroventricularly for 2 wk (\(n = 7–8\)/rats/group) as described in Protocol 1. Under halothane anesthesia, a PE-50 catheter was inserted into the left carotid artery. The next morning, in conscious free-moving rats, the carotid arterial catheters were connected to pressure transducers. After a 30-min rest, BP and HR were recorded for 30 min. A total of 2 ml of blood was then withdrawn from the arterial line and collected into an ice-chilled tube containing EDTA. The animals were then killed by decapitation.

Brain and adrenal tissues were obtained as described previously (37, 38). The hypothalamus was dissected at 4°C according to Glowinski and Iversen (7). OLC was extracted by mixing plasma samples with 1 vol of 0.1% tritiated acetic acid. Tissues were homogenized in 10 vol methanol-2 M acetic acid. The homogenate was centrifuged, and the supernatant was dried by using a vacuum concentrator (model SC110 Speed-Vac, Savant Instruments, Farmingdale, NY), and reconstituted with 0.1% tritiated acetic acid. Plasma and tissue extracts were passed through a 200-mg water-equilibrated Sep-Pak C\(_{18}\) column (Waters). OLC was eluted with 3 ml of 25% acetonitrile. The eluates were dried with the vacuum concentrator, and the extracts were dissolved by using PBS (10 mM, pH 7.4). The anti-ouabain antibody was raised in rabbits immunized with the commercially available cardenolide ouabain conjugated with bovine serum albumin. This antibody has a high antibody titer (1:1.6 \(\times 10^5\)), full cross-reactivity with ouabain, 8% cross-reactivity with digoxin, and minimal cross-reactivity with numerous common endogenous steroids and peptides (38). OLC was measured by ELISA as recently described in detail (37, 38).

#### Protocol 3

By adjusting the amount of NaCl added, aCSF containing 0.14, 0.15, or 0.16 M Na\(^+\) was prepared. After intracerebroventricular cannulation, aldosterone dissolved in aCSF containing 0.14, 0.15, or 0.16 M Na\(^+\), respectively, was infused at 22.5 ng·4.5 \(\mu\)l·h\(^{-1}\)·l\(^{-1}\) for 2 wk in three groups of Dahl S rats (\(n = 7–8\)/group). On day 14 of the infusion, with rats under halothane anesthesia, a PE-50 catheter was placed into the left carotid artery. In the morning of day 15, the resting BP and HR were recorded as described in Protocol 2. The rats were then anesthetized with halothane and placed in a stereotaxic frame. A 23-gauge stainless steel needle was inserted into the cisterna magna.
as described previously (20). A total of 100 μl of CSF was then withdrawn during 1–2 min, and a 1 ml blood sample was taken.

Concentrations of CSF and plasma electrolytes were determined by using ion-selective electrodes (model 917; Hitachi).

Data Analysis

Responses of RSNA were expressed as percentages of resting values. To evaluate the arterial baroreflex function, changes in RSNA/HR (ΔRSNA/ΔHR) in response to changes in MAP were analyzed as a logistic model, using the equation

$$\Delta \text{RSNA}/\Delta \text{HR} = P_1 + P_2[1 + e^{P_3 (\text{MAP} - P_4)}],$$

where $P_1$ is lower ΔRSNA/ΔHR plateau, $P_2$ is ΔRSNA/ΔHR range, $P_3$ is a curvature coefficient, and $P_4$ is MAP50, i.e., the MAP at half the ΔRSNA/ΔHR range. For comparisons of daily water intake, a repeated-measures ANOVA was performed. For comparisons of other data, a two-way ANOVA with the grouping factor was performed. Statistical significance was defined as $P < 0.05$.

RESULTS

In both Dahl S and Dahl R rats, intracerebroventricular infusion of aldosterone at the rate of 22.5 ng/h for 2 wk had no effects on daily water and food intake (not shown), gain of body weight, hematocrit, and plasma Na⁺, K⁺, and Cl⁻ concentrations (Table 1).

Protocol 1

Dahl S rats treated with aldosterone for 2 wk demonstrated significantly higher resting BP and HR compared with Dahl S rats treated with vehicle (131 ± 3 vs. 108 ± 2 mmHg; and 513 ± 19 vs. 467 ± 18 beats/min; $P < 0.05$, for both). Intracerebroventricular aldosterone had no effects on resting MAP and HR in Dahl R rats (110 ± 2 vs. 107 ± 2 mmHg; and 454 ± 10 vs. 437 ± 10 beats/min).

Responses to air stress and intracerebroventricular guanabenz. Air stress caused mild increases in RSNA, MAP, and HR (Figs. 1 and 2), which were similar in Dahl S and Dahl R rats. Figure 1 shows representative tracings in a Dahl S rat infused with aldosterone or vehicle. In Dahl S rats with intracerebroventricular aldosterone, the magnitudes of increases in RSNA, MAP, and HR were approximately twofold of those in Dahl S rats with vehicle or Dahl R rats with either aldosterone or vehicle. Aldosterone did not affect the responses to air stress in Dahl R rats. When MAP and HR responses were expressed as percentages of basal values, a similar enhancement of the responses was observed in Dahl S rats treated with aldosterone versus Dahl S rats treated with vehicle (MAP: 19 ± 3 vs. 8 ± 2%; HR: 8 ± 0.7 vs. 4 ± 0.2%, $P < 0.05$ for both).

Intracerebroventricular guanabenz decreased RSNA, MAP, and HR in a dose-related manner (Fig. 3). In Dahl R rats, aldosterone treatment did not affect the maximal decreases in RSNA, MAP, and HR in response to intracerebroventricular guanabenz. In contrast, in Dahl S rats treated with aldosterone, maximum decreases in RSNA, MAP, and HR in response to the two doses of guanabenz were approximately twofold of those in Dahl S rats treated with vehicle and Dahl R rats treated with aldosterone or vehicle. The percent changes of MAP and HR were also significantly larger in Dahl S rats treated with aldosterone versus Dahl S rats treated with vehicle (decreases in MAP to 50 μg of guanabenz: 18 ± 2 vs. 10 ± 1%; HR: 6 ± 0.3 vs. 4 ± 0.2%, $P < 0.05$ for both).

Responses to intracerebroventricular ouabain. Intracerebroventricular ouabain increased RSNA, MAP, and HR (Fig. 4). In Dahl R rats, intracerebroventricular treatment with aldoste-

![Fig. 2. Maximal increases in MAP, RSNA, and HR in response to air-jet stress in DS and DR rats after icv infusion of Aldo or Veh [artificial cerebrospinal fluid (aCSF) with 0.15 M Na⁺] for 2 wk. Data are means ± SE (n = 6–8). *P < 0.05 vs. others.](http://alp.ajpheart.org/)

![Fig. 1. Analog recording of blood pressure (BP), heart rate (HR), and integrated renal sympathetic nerve activity (RSNA) in response to air-jet stress in a Dahl salt-sensitive (DS) rat after intracerebroventricular (icv) infusion of aldosterone (Aldo; A) or vehicle (Veh; B) for 2 wk. bpm, Beats/min.](http://alp.ajpheart.org/)
Table 2. Effects of intracerebrovascular infusion of Aldo for 2 wk on baroreflex control of RSNA and heart rate in Dahl S and R rats

<table>
<thead>
<tr>
<th>Protocol</th>
<th>Dahl R</th>
<th>Dahl S</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vehicle</td>
<td>Aldo</td>
<td>Vehicle</td>
</tr>
<tr>
<td>No. of Rats</td>
<td>6</td>
<td>8</td>
</tr>
<tr>
<td>RSNA</td>
<td></td>
<td></td>
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<tr>
<td>P2, range, %</td>
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<td>180±14</td>
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<td>Average slope, %/mmHg</td>
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<td>−2.84±0.12</td>
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<td>Pe, MAP50, mmHg</td>
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<td>109±3</td>
</tr>
<tr>
<td>Heart Rate</td>
<td></td>
<td></td>
</tr>
<tr>
<td>P2, range, beats/min</td>
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<td>226±17</td>
</tr>
<tr>
<td>Average slope, beats·min⁻¹·mmHg⁻¹</td>
<td>−2.13±0.19</td>
<td>−1.95±0.17</td>
</tr>
<tr>
<td>Pe, MAP50, mmHg</td>
<td>106±3</td>
<td>108±3</td>
</tr>
</tbody>
</table>

Values are means ± SE estimated by logistic analysis. P2, change in renal sympathetic nerve activity (RSNA)/change in heart rate range; Pe, mean arterial pressure at half the change (MAP50) in RSNA/change in heart rate range. *P < 0.05, vs. others.

Protocol 2

In this study protocol, Dahl S rats treated with aldosterone for 2 wk also demonstrated significantly higher resting BP and HR compared with Dahl S rats treated with vehicle (125 ± 2 vs. 106 ± 2 mmHg; and 499 ± 15 vs. 453 ± 13 beats/min, P < 0.05 for both). Aldosterone had, again, no effects on resting MAP and HR in Dahl R rats (105 ± 3 vs. 103 ± 3 mmHg; and 451 ± 12 vs. 435 ± 19 beats/min).

In Dahl R rats, chronic intracerebrovascular infusion of aldosterone had no effects on OLC content in the hypotalamus, plasma, or adrenal gland. In Dahl S rats, chronic intracerebrovascular aldosterone increased hypothalamic OLC by ~70%, without parallel increases in peripheral OLC (Fig. 6).

Protocol 3

Intracerebrovascular aldosterone in 0.15 M Na⁺ caused a similar increase in resting MAP and HR in Dahl S rats as in protocol 2 [131 ± 3 vs. 125 ± 2 mmHg; 490 ± 10 vs. 499 ± 15 beats/min, not significant (NS) for both] (Fig. 7). Resting MAP and HR in Dahl S rats treated with intracerebrovascular aldosterone in 0.14 M Na⁺ were significantly lower than those rats treated with aldosterone in 0.15 or 0.16 M Na⁺ and were similar to Dahl S rats infused with vehicle alone in protocol 2 (111 ± 3 vs. 106 ± 2 mmHg; 465 ± 10 vs. 453 ± 13 beats/min, NS for both). Intracerebrovascular aldosterone in 0.16 M Na⁺ caused the largest increase in resting MAP (147 ± 3 mmHg, P < 0.05 vs. others). CSF [Na⁺] in the cisterna magna showed a minor, nonsignificant increase: 147.2 ± 1.7 vs. 146.3 ± 1.5 mM for aCSF with 0.16 vs. 0.14 M Na⁺. CSF K⁺ and Cl⁻ concentrations were similar among three groups (2.7 ± 0.1, 2.7 ± 0.2, and 2.6 ± 0.1 mM and 118 ± 2, 117 ± 1, and 117 ± 2 mM, respectively, for rats with intracerebrovascular aCSF containing 0.14, 0.15, and 0.16 M Na⁺). Plasma electrolyte concentrations were similar among these three groups of rats (not shown).

DISCUSSION

The present study demonstrates as new findings that 1) in Dahl S rats, chronic intracerebrovascular infusion of aldosterone...
Aldosterone in aCSF with [Na\(^+\)] slightly higher than CSF significantly increases resting MAP associated with sympathetic hyperreactivity and impairment of arterial baroreflex control of RSNA and HR as well as a clear increase in OLC content in the hypothalamus; 2) the extent of the pressor effect of intracerebroventricular aldosterone appears to depend on the [Na\(^+\)] in the aCSF vehicle; and 3) in Dahl R rats, chronic intracerebroventricular infusion of aldosterone has no effects on sympathetic reactivity, BP, arterial baroreflex function, and brain OLC content.

Intracerebroventricular infusion of aldosterone dissolved in vehicle with 0.14 M Na\(^+\) did not affect resting BP and HR in Dahl S rats, but aldosterone dissolved in vehicle containing 0.15 or 0.16 M Na\(^+\) markedly increased BP and HR. The pressor response to aldosterone in vehicle with 0.16 M Na\(^+\) was significantly larger than that to aldosterone in vehicle with 0.15 M Na\(^+\). These results are consistent with findings in Wistar rats (36) showing that acute intracerebroventricular aldosterone plus intracerebroventricular infusion of aCSF with 0.16 M Na\(^+\) but not with regular aCSF (with 0.145 M Na\(^+\)) increases resting RSNA, BP, and HR. In the present study, the aCSF was infused into one of the lateral ventricles. The CSF sampled through the cisterna magna showed only a minor (1 mM) increase in [Na\(^+\)] by intracerebroventricular infusion of aCSF with 0.16 vs. 0.14 M Na\(^+\). However, it is likely that the [Na\(^+\)] in the CSF at local areas near the infusion site in the forebrain was higher. Increases in CSF [Na\(^+\)] by only 2–3 mM can significantly increase the firing rate of neurons in the paraventricular or supraoptic nucleus (16), and such local increases may be sufficient to explain the different responses in brain OLC, sympathetic reactivity, and BP to intracerebroventricular aldosterone in aCSF with 0.16 vs. 0.14 M Na\(^+\).

Intracerebroventricular aldosterone increased OLC content in the hypothalamus in Dahl S rats. The actual cellular mechanisms leading to increases in OLC by aldosterone have not yet been addressed. One possibility is that intracerebroventricular aldosterone activates MR and ENaC located in the ventricular ependyma (Refs. 33, 35, and 39) and thereby increases the entry of CSF Na\(^+\) through these channels into the interstitial fluid surrounding neurons and glia, which in turn increases intracellular [Na\(^+\)] and Ca\(^{2+}\) concentration ([Ca\(^{2+}\)]) of ouabain-producing cells, such as astrocytes (23) and/or the neurosecretory neurons of the paraventricular nucleus (34), leading to increase in OLC synthesis and/or release. Whether or not aldosterone also directly increases Na\(^+\) entry into neurons or glia has, to our knowledge, not yet been studied. Sympathoexcitatory and pressor responses to intracerebroventricular injection of exogenous ouabain became markedly smaller in Dahl S rats after intracerebroventricular aldosterone. These findings suggest that intracerebroventricular aldosterone not
only increases brain OLC content but also OLC release, leading to increased receptor occupancy and thereby decreased availability of free receptors and attenuated responses to exogenous ouabain.

Sympathetic hyperactivity has been proposed as one of the central mechanisms mediating intracerebroventricular aldosterone-induced hypertension (4, 9). Consistent with our previous study in Wistar rats (36), the present study shows that in Dahl S rats, chronic intracerebroventricular infusion of aldosterone clearly affects sympathetic reactivity. Intracerebroventricular aldosterone enhanced sympathoexcitatory and pressor responses to air stress, indicating an increased activity of central sympathoexcitatory pathways. Intracerebroventricular aldosterone also enhanced sympathoinhibitory and depressor responses to intracerebroventricular guanabenz, consistent with a decreased activity in central sympathoinhibitory pathways (25, 30). Moreover, intracerebroventricular aldosterone markedly impaired arterial baroreflex control of RSNA and HR in Dahl S rats. This impairment is likely due to the aldosterone-induced activation of brain OLC and the brain RAS (17). Although intracerebroventricular aldosterone at 22.5 ng/h unlikely has direct peripheral effects (31, 35), one cannot exclude indirect peripheral mechanisms contributing to baroreflex desensitization. In the present study, no parameter of resting sympathetic activity per se was measured. The pattern of changes discussed above is consistent with an increase in resting sympathetic activity, but this needs to be substantiated in further studies.

Increased brain OLC and subsequent increase in activity of brain RAS may also increase the synthesis/release of vasopressin (29), and the latter may contribute to the increase in BP. However, intracerebroventricular aldosterone at 25–28 ng/h for 2 wk has no effects on parameters, such as hematocrit, plasma electrolytes, plasma concentrations of renin, vasopressin or aldosterone, or water intake in Sprague Dawley (31) or Wistar (the present study and Ref. 36) rats. Intracerebroventricular aldosterone induces hypertension even in rats fed a Na\textsuperscript{+}-deficient diet (31). In addition, increased plasma vasopressin may sensitize the arterial baroreflex (14). The opposite was observed in the present study.

Compared with previous studies in Sprague-Dawley (21) and Wistar (36) rats, the increases in hypothalamic OLC (36) and in BP (31, 36) by intracerebroventricular aldosterone are
twofold larger in Dahl S rats. These results suggest that Dahl S rats exhibit an upregulation or gain of function of brain MR per se or increased responsiveness of mechanisms downstream to the MRs, e.g., benzamil-blockable ENaCs or OLC-producing systems. On the other hand, in Dahl R rats, intracerebroventricular infusion of aldosterone does not increase hypothalamic OLC and neither causes sympathetic hyperactivity and hypertension. Thus loss of function of brain MRs to aldosterone or of Na\(^+\) channels to MR stimulation may contribute to the salt-resistant nature of Dahl R rats. In Dahl S and Dahl R rats, neither aldosterone metabolism, MR characteristics in the brain, nor changes in brain Na\(^+\) channels by MR activation have been studied. Chronic intracerebroventricular infusion of aldosterone in regular aCSF had no effects on resting BP in sheep (28). It is possible that sheep may also respond to intracerebroventricular aldosterone if combined with a small increase in vehicle [Na\(^+\)]. Alternatively, sheep may resemble Dahl R rats in their responses to central aldosterone.

In Dahl S rats, intracerebroventricular infusion of aldosterone elicits the same central nervous system (CNS) phenotype as caused by high dietary salt (1, 17, 27), including increased brain OLC and decreased responses to intracerebroventricular injection of ouabain as well as sympathetic hyperreactivity, impairment of baroreflex function, and hypertension. Blockade of brain MR prevents the hypertension in Dahl S rats on high salt (10). Intracerebroventricular infusion of a MR antagonist also prevents impairment of baroreflex function and hypertension in spontaneously hypertensive rats (32) and DOCA-salt rats (22). These findings suggest that effects of aldosterone in the CNS contribute to high salt-induced sympathetic hyperactivity and hypertension. High salt intake decreases plasma aldosterone levels (15). So far, no studies have assessed whether high salt intake increases levels of aldosterone or other MR agonists in relevant brain regions of Dahl S rats. Figure 8 provides a schematic outline of the cascade of CNS mechanisms that may be activated by high salt intake and MR stimulation in Dahl S and not Dahl R rats.

**Possible Limitation of This Study.**

The RSNA signals deteriorate gradually during the first postoperative day, and the studies were therefore performed ≥4 h after the rat had recovered from anesthesia. Although resting BP was similar in protocol 1 (≥4 h after an extensive surgery) versus protocol 2 values (≥24 h after a minor surgery), in both protocols, the resting HR was 50–60 beats/min higher compared with those values obtained through radiotelemetry (19). Therefore, postoperative stress may likely still contribute to sympathoexcitation, and higher resting values of BP and HR in Dahl S versus Dahl R rats may partly be due to higher responsiveness to stress in Dahl S rats.

In summary, the present study demonstrates that in Dahl S but not Dahl R rats, chronic intracerebroventricular infusion of aldosterone in aCSF containing Na\(^+\) that is just above the physiological concentration causes sympathetic hyperreactivity and hypertension and impairment of arterial baroreflex control of RSNA and HR. The pressor effect of intracerebroventricular aldosterone is absent when the [Na\(^+\)] of the vehicle is lower than regular CSF and magnified when the [Na\(^+\)] is increased. The sympathetic hyperreactivity and hypertension by intracerebroventricular aldosterone are associated with significant increases in OLC content in the hypothalamus in Dahl S rats. Unlike Wistar and Dahl S rats, in Dahl R rats, intracerebroventricular aldosterone does not affect brain OLC, sympathetic reactivity, and BP, suggesting that in this rat strain loss of function of brain MRs or mechanisms downstream to MR activation may contribute to its salt-resistant nature.

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**REFERENCES**


