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Sodium Excretion after Adrenal Enucleation and during Adrenal Regeneration Hypertension (40870)

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Abstract. Renal function following saline infusion (10% body wt) has been studied in four groups of rats 3 to 5 days after operation: Sham (S), Adrenal Enucleation (AE), Uninephrectomy (N), and AE + N. The purpose of the study was to clarify the mechanisms of the effect of N on sodium excretion in AE. AE excreted significantly less sodium, 6.7 ± 1.3 μEq/min, than S, 29.7 ± 2.7 μEq/min. Sodium excretion by N, 25.4 ± 5.3 μEq/min and AEN, 19.4 ± 3.2 μEq/min were not different from each other or S, but greater than AE ($P < 0.01$). When AE (two kidneys) was compared with AEN (one kidney), the filtered sodium was not significantly different. Thus, it is concluded that the greater natriuresis in AEN versus AE is most likely secondary to a difference in tubular sodium reabsorption. Renal function following saline infusion (10% body wt) has been studied after the development of adrenal regeneration hypertension (6 weeks postsurgery) in AEN rats and their control groups N. Mean blood pressure was significantly elevated in the AEN group, 161 ± 4 versus 128 ± 3 mmHg. The sodium excretion was not different between the two groups. It is concluded that rats with adrenal regeneration hypertension do not have an exaggerated natriuresis as has been described in other forms of experimental hypertension.

Adrenal regeneration hypertension (ARH) develops in rats following removal of the adrenal glands without their capsules (so-called adrenal enucleation (AE)), uninephrectomy, and the ingestion of saline in place of drinking water (1, 2). Several years after the discovery of this phenomenon, Gaunt and associates (3, 4) noted that during the first few days after AE, rats were unable to excrete an acute sodium load. They postulated therefore that the impaired natriuresis might be causally related to the development of ARH. Recently Hall *et al.* (5, 6) confirmed that after AE, rats had an impaired natriuresis but that this could be corrected by the removal of one kidney in these AE rats. The mechanism of this natriuretic effect of uninephrectomy has not been investigated. Specifically, Hall *et al.* (6) did not evaluate renal function.

The purpose of our studies was to clarify the mechanism of this phenomenon described by Hall *et al.* (5, 6) by evaluating renal hemodynamics during acute saline loading. In addition, we have extended these studies to measure sodium excretion after the onset of hypertension to determine whether rats with presumed mineralocorticoid hypertension have an exaggerated natriuresis as has been described in other forms of hypertension (7–10).

Materials and methods. Thirty-six Charles River CD rats weighing 270–330 g were anesthetized with sodium brevital, 70–75 mg/kg body wt. Through a flank incision, both adrenal glands were exposed and lifted out of the incision without impairment of the blood supply. In 18 animals, the glands were replaced in the suprarenal area. In 6 of these 18 animals nothing further was done (Sham, S), while in the other 12 animals a right nephrectomy was performed (Uninephrectomy, N). In the remaining 18 animals, both adrenal capsules were slit, the glands gently squeezed out with a small forceps and the capsules replaced in the suprarenal area. In 6 of these animals (adrenal enucleation, AE) nothing further was done, while in the other 12 animals the right kidney was also removed (adrenal enucleation uninephrectomy, AEN). After recovery from surgery and anesthesia, all of the animals were allowed free access to food

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and were provided 1% saline as drinking water.

Clearance studies. Twenty-four rats, six from each of the four groups described above, were studied 2 to 6 days after surgery, while six N and six AEN rats were maintained on saline for 6 weeks and then studied. The animals were anesthetized by intraperitoneal injection of ketamine hydrochloride, 8.5 mg/100 g body wt, with supplemental small doses (1/5 of the original dose) as needed. After tracheostomy, polyethylene catheters were placed in the jugular veins for infusions. A femoral artery was catheterized for blood sampling and blood pressure recordings. A catheter was placed in the bladder through an abdominal wall incision, and urine collected in weighed tubes. Fifteen μCi/hr of [*carboxy*-^{14}C]inulin (New England Nuclear Corp.) in saline was infused at a constant rate of 0.03 ml/min. After 1 hr of inulin infusion, a volume of 0.9% saline equal to 10% body wt was infused over 1 hr, and the rate of infusion was then reduced by one-half. Thirty minutes after slowing the saline infusion, urine collections were obtained every 20 min with accompanying mid-point blood sample. The clearance data presented represent the mean of three to four 20-min collections. It was not possible to obtain adequate clearance measurements in the AE and AEN rats prior to saline expansion because of the extremely low urine flow rate.

Plasma and urine radioactivity was determined by liquid scintillation counting in Aquasol (New England Nuclear, Boston, Mass.) The minimum total counts were 30× background. Plasma and urine sodium concentrations were determined with a flame photometer (Instrumentation Laboratory, Inc., Lexington, Mass.).

Standard statistical techniques were used and results are presented as means ± SE. Comparison of means among groups was performed by analysis of variance and Duncan's multiple range test or the unpaired *t* test as appropriate (11).

Results. The renal and systemic parameters measured in the four groups studied 2 to 6 days after operation are summarized in Table I.

No differences were noted among groups in body weight, mean blood pressure, hematocrit, or plasma sodium or potassium concentration. Glomerular filtration rate per kidney increased as expected after uninephrectomy, 51% N vs S and 74% AEN vs AE. Absolute sodium excretion was reduced in AE rats, compared to S; 29.7 ± 2.7 vs 6.7 ± 1.3 μEq/min ($P < 0.001$). However, sodium excretion in the AEN group, 19.4 ± 3.2 μEq/min, was not significantly different from its control group N, 25.4 ± 5.3 μEq/min. Of greatest interest was the comparison between the AE and AEN groups. There was no significant difference in the amount of sodium filtered by the AE group with two kidneys, 292 ± 29 μEq/min,

TABLE I. RENAL AND SYSTEMIC PARAMETERS AFTER ADRENAL ENUCLEATION[a]

	S	AE	N	AEN
Number of rats	6	6	6	6
Body weight (g)	302 ± 10	285 ± 8	298 ± 5	289 ± 4
GFR (ml/min)	2.94 ± 0.26	1.87 ± 0.19*	2.1 ± 0.20*	1.63 ± 0.12*[b]
Filtered sodium (μmole/min)	463 ± 41	292 ± 29*	351 ± 38*	246 ± 22[b]
Sodium excretion (μmole/min)	29.7 ± 2.7	6.7 ± 1.3*	25.4 ± 5.3	19.4 ± 3.2[c]
Fractional sodium excretion (%)	6.7 ± 0.8	2.3 ± 0.3*	7.1 ± 1.1	7.8 ± 1.0[c]
Blood pressure (mmHg)	111 ± 2	116 ± 3	121 ± 8	112 ± 3
Plasma sodium (mmole/liter)	158 ± 1	156 ± 2	158 ± 5	150 ± 4
Plasma potassium (mmole/liter)	4.8 ± 0.3	4.2 ± 0.2	4.3 ± 0.2	4.0 ± 0.1
Hematocrit (%)	39 ± 2	38 ± 1	40 ± 1	41 ± 1

[a] S = sham, AE = adrenal enucleation, N = uninephrectomy, AEN = adrenal enucleation uninephrectomy.
[b] Versus N.
[c] Versus AE.
* $P < 0.01$ vs S.

or the AEN group with one kidney, 246 ± 22 μEq/min. However, both absolute sodium excretion and fractional sodium excretion were significantly larger in the AEN rats.

The renal and systemic parameters for the AEN and N groups studied 6 weeks after operation are summarized in Table II. BP was significantly elevated in the AEN group, 161 ± 4 vs 128 ± 3 mmHg. Among all other parameters studied, no significant differences were noted between these two groups. In particular, GFR was no longer different. Absolute Na excretion was significantly increased compared to the AEN and N groups studied shortly after surgery while fractional excretion was not different.

Discussion. It is clear from the data of Gaunt et al (3, 4) and from our own laboratory (12) that AE impairs natriuresis in response to acute saline loading. In our previous studies we have demonstrated that this antinatriuresis was secondary to a mineralocorticoid enhanced tubular reabsorption of sodium along the collecting duct (13). Hall *et al.* (6), but not Paulson and Eversole (14), found that uninephrectomy normalized the natriuretic response to an oral saline load in AE rats but the mechanism of this phenomenon was not studied. We have previously demonstrated that the antinatriuresis of AE can be reversed by increasing the amount of sodium filtered, e.g., by dexamethasone (13) or by inhibiting the action of the mineralocorticoid responsible for the antinatriuresis with spironolactone (13). Thus, there were at least two possibilities to explain the effect of uninephrectomy in AE rats, an increase in filtered sodium because of an increase in GFR or an inhibition of tubular sodium reabsorption.

We found no significant difference in the amount of sodium filtered by the AE group with two kidneys or the AEN group with one kidney. Since sodium excretion was almost three times greater in the AEN rats, we believe these data provide strong evidence that the enhanced natriuresis which is found during volume expansion of uninephrectomized AE rats occurs because of a decrease in tubular reabsorption of sodium and not because of an increased filtered load.

Wilson and Sonnenberg (15) have recently demonstrated that there is a decrease in medullary collecting duct sodium reabsorption after a reduction in renal mass and volume expansion. We previously found that the antinatriuresis of AE occurs along the collecting duct (12, 13). It seems reasonable to postulate that the reduction in renal mass negates the mineralocorticoid stimulated increase in tubular sodium reabsorption during volume expansion, and that this effect probably occurs along the collecting duct.

The finding of a normal natriuresis in AEN rats cannot be cited as evidence against the secretion of a salt-retaining factor being produced by the adrenal capsule. In daily balance studies we have found that both AE and AEN rats are in significant positive sodium balance (16). Thus, it appears that the natriuretic effect of acute volume expansion in the uninephrec-

TABLE II. RENAL AND SYSTEMIC PARAMETERS DURING ADRENAL REGENERATION HYPERTENSION[a]

	N	AEN
Number of rats	6	6
Body weight (g)	350 ± 6	364 ± 13
GFR (ml/min)	3.27 ± 0.2	3.18 ± 0.3
Sodium excretion (μmole/min)	41.7 ± 1.5	34.5 ± 3.4
Fractional sodium excretion (%)	8.3 ± 0.5	7.0 ± 0.6
Blood pressure (mmHg)	128 ± 3	161 ± 4*
Plasma sodium (mmole/liter)	156 ± 1	157 ± 1
Plasma potassium (mmole/liter)	4.2 ± 0.2	4.3 ± 0.1
Hematocrit (%)	43 ± 1	45 ± 1

[a] N and AEN as in Table I.
* $P < 0.01$.

tomized AE rat can overcome the antinatriuretic state after AE when two kidneys are present.

The data obtained after the onset of ARH are also of interest since the response of ARH rats to saline loading has not been previously reported. There is a large body of data demonstrating that hypertensive man and animals exhibit an exaggerated natriuresis when challenged with a salt load. This exaggerated natriuresis has been demonstrated in man (17, 18) and in rats with Goldblatt hypertension (10) and in two rat strains with genetic susceptibility to hypertension (7, 8, 9), as well as DOC-salt hypertension[2] (19). While it is appreciated that not all investigators have found an exaggerated natriuresis in the SHR strain (20, 21) one study in this strain is of particular interest with reference to the present data. Willis and Bauer (8) evaluated the importance of aldosterone in the exaggerated natriuresis seen in SHR rats. They found that treatment with aldosterone eliminated this increased natriuresis in the SHR group while spironolactone increased sodium excretion in normal rats to equal that of the SHR group. If their data can be extended to ARH, it is tempting to propose a possible explanation for the lack of an exaggerated natriuresis in our studies. ARH is thought to be a steroid mediated hypertension (22–24). It may be that the increase in mineralocorticoid prevents the exaggerated natriuresis in these animals.

In summary, we have found that uninephrectomy corrects the impaired natriuretic response of AE rats. We have furthermore shown that AEN rats who develop ARH do not have an exaggerated natriuresis after saline expansion. We conclude that the natriuretic effect of uninephrectomy on AE rats described by Hall *et al.* (6) is consistent with an effect of uninephrectomy on tubular sodium reabsorption. The lack of an exaggerated natriuresis in ARH may be related to an increase in mineralocorticoid activity.

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