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The Effect of Sodium Depletion on the Renal Response to Short-Duration NH_4Cl Acid Loading (39719)

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Introduction. Current concepts of the renal control of external hydrogen ion balance emphasize the importance of the relationships between the delivery of sodium to the distal nephron, the avidity of the distal exchange sites for sodium, and the excretion of poorly permeant anions (1). Sodium depletion would be expected to have two independent and opposing influences on renal acidification. Although the distal delivery of sodium might decline, distal sodium for hydrogen exchange would presumably be facilitated (2). There have been very few systematic studies of the influence of sodium depletion on renal acidification. To our knowledge the only study in humans in which volunteers were acid loaded both prior to and after sodium depletion showed an impairment in ammonium excretion (without change in urine pH) in the latter case (3). On the other hand, Schwartz *et al.* (4) found that sodium-depleted acid-loaded subjects excreted ammonium at a rate greater than that reported by others for normal sodium-replete subjects. We present here our findings in five normal subjects who underwent short-duration NH_4Cl acid-loading tests both before and after the induction of mild sodium depletion. The results suggest that sodium depletion limits maximal distal hydrogen ion secretion in response to acid loading.

Materials and methods. Five healthy male volunteers aged 20–25 years, hospitalized at the Clinical Research Center, University of Miami School of Medicine were evaluated. None gave a history of renal or cardiovascular disease or was receiving any medications. Creatinine clearance and serum sodium, potassium, chloride, and bicarbonate concentrations were normal (Table I). There was no glucosuria or proteinuria and there were no abnormalities in the urinary sediment. Urine cultures showed no growth in all subjects.

After admission the subjects were given a diet containing 150 mEq of sodium and 60 mEq of potassium daily for 3 days. On Day 4 the subjects underwent a short-duration NH_4Cl acid-loading test. Immediately after the acid-loading test the patients were started on a 10-mEq sodium, 60-mEq potassium diet until Day 8 when the NH_4Cl test was repeated. A single dose of furosemide, 20 mg by mouth, was given the morning following the first acid-loading test. The subjects had access to unrestricted amounts of distilled water; daily activities were not limited. Body weight, blood pressure sitting and standing, and 24-hr urinary sodium, potassium, and creatinine excretion were measured daily throughout the study.

Acid loading. The evening prior to the study, the subject ate his usual meal and then had no food until the protocol was completed, but was allowed water in small amounts. He remained recumbent as much as possible after awakening on the day of the study. All urines were obtained by spontaneous voiding and collected under mineral oil. At 7:00 and 8:00 AM (baseline specimen) the subject voided. At 8:00 AM arterial and venous blood samples were drawn for baseline measurements. In obtaining the venous sample, blood for serum determinations was drawn first into a 20-ml syringe with the tourniquet being released immediately to avoid stasis. A 2-ml heparinized syringe was then transferred to the needle for aspiration of the blood sample for pH and PCo_2 measurements. The subject was then given 1.9 mEq/kg body weight of NH_4Cl prepared in gelatin capsules and ingested with a total of 200 ml of water over

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TABLE I. EFFECT OF SODIUM DEPLETION ON SERUM AND URINARY ELECTROLYTES.

	Weight (kg)	$U_{Na}V$[a] (μEq/min)	U_KV[a] (μEq/min)	4-Day cumulative urinary balance (mEq) Na	K	Serum Na	K	HCO_3 (mEq/liter)	Cl	Ccr[b] (ml/min)
Before Na depletion	78.3±6.3[c] (69-101)	161±5 (153-179)	63±6 (42-77)	–	–	140±1 (138-141)	4.4±0.2 (3.9-4.9)	27±2 (21-30)	104±1 (100-107)	138±9 (121-171)
P value (n = 5)	<0.001	<0.001	NS[d]	–	–	NS	NS	NS	<0.05	<0.025
After Na depletion	76.6±6.4	7±3	57±5	−213±34	+11±8	138±1	4.2±0.1	28±1	101±1	125±12
	(67-99)	(1-16)	(43-70)	(−135--323)	(−5-+32)	(135-143)	(3.6-4.4)	(25-31)	(100-105)	(103-170)

[a] A 24-hr period preceding acid loading.
[b] Average of three to five periods during acid loading.
[c] Mean ± SE. Ranges are within parentheses.
[d] NS, not significant ($P > 0.05$).

approximately a 2-hr period. Urines were obtained hourly for 6 hr after ingestion of the acid load. Venous blood was drawn 2 hr and again 6 hr after the acid load. Fifty milliliters of water per hour were given by mouth and sodium-free candy provided as desired during the study.

Written consent was obtained after explanation of the details of the procedures and the potential risks. The research was carried out according to the principles outlined in the Declaration of Helsinki and was approved by the Human Experimentation Committees of the University of Miami and the Miami Veterans Administration Hospital. No adverse effects occurred.

Determination of the pH and PCo_2 of blood and urine was completed within 5 min of collection on a Radiometer Acid-Base Cart ABC 1. All urine and venous blood samples were measured for concentrations of sodium, potassium, phosphorus, creatinine, and total solutes. Urinary titratable acidity (TA) and ammonium (NH_4) content were also measured. None of the urine specimens tested contained glucose or abnormal amounts of protein. The methods and calculations employed are those previously described from this laboratory (5, 6). The data are presented as means ± SE and were evaluated statistically by means of Student's t test for paired values (7).

Results. Achievement of sodium depletion (Table I). As a result of the sodium-restricted diet, the initial NH_4Cl acid loading (3), and furosemide administration, each of the five patients developed mild to moderate sodium depletion. This was manifested by an average weight loss of 1.7 ± 0.1 (SE) kg, cumulative urinary sodium loss of 213 ± 13 mEq up until the day prior to the second acid-loading test, the excretion of 7 ± 3 mEq of sodium during the 24-hr period prior to restudy, and a 9% decrease in endogenous creatinine clearance. Urinary potassium balance was unchanged. The serum concentrations of Na, K, and HCO_3 did not change significantly as a result of sodium depletion. The serum chloride concentration was slightly but significantly lower after sodium depletion.

Response to acid loading (Table II). There were no significant differences between the baseline arterial pH (before, 7.40 ± 0.01; after, 7.42 ± 0.02) or venous pH (before, 7.31 ± 0.02; after, 7.31 ± 0.01), or arterial HCO_3 (before, 25 ± 1; after, 28 ± 1 mEq/liter) or venous HCO_3 (before, 27 ± 2; after 28 ± 1 mEq/liter) values prior to and following sodium depletion. On both occasions acid loading resulted in significant and approximately equivalent decreases in venous pH (minimum before, 7.25 ± 0.01; after, 7.27 ± 0.01) and HCO_3 (minimum before 22 ± 1; after 22 ± 1 mEq/liter).

After sodium depletion minimal urine pH was higher (average increment 0.43 ± 0.07; range, 0.26 to 0.62; $P < 0.001$) in each subject, and maximal $U_{NH4}V$ was lower ($P < 0.02$) following NH_4Cl acid loading (Table

TABLE II. URINARY CHANGES FOLLOWING ACID LOADING.

Patient		UpH[a]	$U_{NH_4}V$[a] (μEq/min)	$U_{NH_4}V$[b] (μEq/min)	$U_{TA}V$[a] (μEq/min)	$U_{Na}V$[c,d] (μEq/min)	U_KV[d] (μEq/min)	UpV[d] (μg/min)	V[d] (ml/min)
1	B[e]	4.60	67	50	–	308	92	730	1.8
	A[f]	5.00	47	41	–	30	66	530	0.9
2	B	4.65	50	46	33	260	114	410	2.3
	A	5.21	39	28	–	12	48	270	1.2
3	B	4.61	47	36	33	253	109	720	2.9
	A	4.90	46	44	31	109	96	490	1.4
4	B	4.79	78	58	38	206	121	920	1.4
	A	5.05	67	46	43	79	167	730	1.0
5	B	4.59	67	46	35	306	114	860	1.9
	A	5.21	47	38	25	28	124	490	0.9
Mean ± SE	B	4.65 ± 0.04	62 ± 6	47 ± 3	35 ± 1	297 ± 19	110 ± 5	728 ± 88	2 ± 0.3
P Value		<0.005	<0.02	NS[g]	NS	<0.005	NS	<0.01	<0.01
	A	5.07 ± 0.06	49 ± 5	39 ± 3	33 ± 5	32 ± 12	100 ± 21	502 ± 73	1 ± 0.1

[a] Maximal or minimal values.
[b] Six-hour average after NH_4Cl.
[c] The relatively elevated $U_{Na}V$ in some patients most likely reflects the natriuretic effect of NH_4Cl (3).
[d] At the time of minimal UpH.
[e] B, before.
[f] A, after.
[g] NS, not significant ($P > 0.05$).

II). Although the average $U_{NH4}V$ for the 6 hr following acid loading was also lower, this difference did not achieve statistical significance. Because of a technical problem, titratable acid excretion rates were available in three subjects only and were not different before or after sodium depletion.

Figure 1 contrasts the increments in minimal urine pH associated with sodium depletion obtained in the present study (open circles) with the variable changes found in 21 repeated acid-loading studies performed in our laboratory in normal subjects and in patients with the incomplete syndrome of distal renal tubular acidosis (closed circles) restudied without changing the dietary sodium intake. In contrast with the mean increment in urine pH of 0.43 induced by sodium depletion, the mean change in minimal urine pH in the other group was −0.003 (range, +0.33 to −0.30).

At the time of minimal pH, $U_{Na}V$, U_pV, and V were significantly lower after sodium deprivation, while U_KV was similar (Table II). Likewise, there were no differences in urine osmolality (before, 677 ± 68 mOsm/kg of H_2O; after, 625 ± 91 mOsm/kg of H_2O). The average $U_{Na}V$ for the 6 hr of the acid-loading study was lower ($P < 0.01$) and the percentage of tubular reabsorption of phosphorus at the time of minimal urine pH was higher ($P < 0.05$) after sodium depletion (before sodium depletion, 195 ± 38

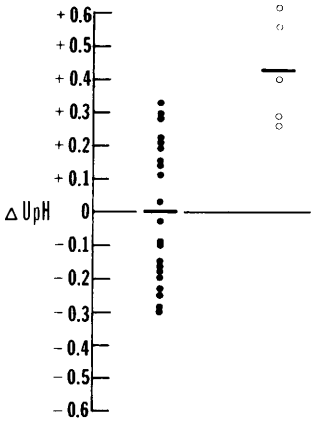

FIG. 1. Changes in minimal urine pH in repeated acid-loading studies. Minimal urine pH was higher (average increment, 0.43 ± 0.07) in each of the five subjects reevaluated during sodium depletion (open circles). Of 21 repeated short-duration acid-loading tests obtained in normal volunteers and in patients with the incomplete syndrome of distal renal tubular acidosis (closed circles), 10 demonstrated increases and 11 demonstrated decreases in minimal urine pH (average change, −0.003 ± 0.05; $P < 0.01$ in comparison to sodium depletion data).

μEq/min and 85.6 ± 0.7%, respectively; after sodium depletion, 18 ± 8 μEq/min and 88.4 ± 0.8%, respectively). The average 6-hr phosphorus and potassium excretion rates did not differ at the time of the two studies.

When $U_{NH4}V$ was plotted against concom-

itant urine pH before and after sodium depletion (Fig. 2), there was considerable overlap of the points. Calculation of the equations for regression lines revealed slopes which differed significantly from zero but not from each other, indicating that for a given change in urine pH the change in $U_{NH4}V$ did not differ in the two studies. In addition, the lines did not differ from each other in elevation.

Discussion. In the present study the average minimal urine pH was higher and the maximal rate of ammonium excretion was lower after modest sodium depletion was induced in five normal volunteers. Although the changes noted were quantitatively small these observations support the concept that alterations in sodium metabolism may affect urinary acidification (1).

The higher minimal urine pH during sodium depletion could not be explained by changes in blood acid-base status. Urine flow rate and urinary phosphorus excretion were both lower following sodium depletion but these changes would not be expected to elevate urine pH (8, 9). In our laboratory, the minimal urine pH after short-duration acid loading has a good reproducibility and in general is likely to be either slightly greater or less than the initial value (average change in urine pH, -0.003 ± 0.05), in marked contrast to the present study in which all five subjects had increments in minimal pH of 0.26 or greater (Fig. 1).

In addition to an increase in minimal urine pH, sodium depletion was associated with a decrease in ammonium excretion. This change may have been due largely to changes in urine pH, since, when $U_{NH4}V$ was plotted against urine pH, the equations for the regression lines were not different either in slope or elevation (Fig. 2).

Potassium loading is known to decrease urinary ammonium excretion (10). Our subjects manifested comparable serum potassium concentrations and urinary potassium excretion rates during the two acid-loading studies, and cumulative urinary potassium balance in between the studies was close to zero, suggesting that important changes in potassium status did not occur during the sodium-depleted state.

Aldosterone secretion is enhanced during sodium depletion and this might be expected to facilitate renal acid excretion (11, 12). It is known, however, that the increased acid excretion often associated with states of mineralocorticoid excess can be prevented by dietary sodium restriction (13). Moreover, limitation of sodium delivery to the distal tubule might be expected to result in a decreased rate of sodium reabsorption in the distal nephron, thus decreasing the negative potential necessary to promote hydrogen ion secretion (14).

Although renal tubular hydrogen ion secretion may be linked somehow to sodium reabsorption, the relationship is not believed to be the result of direct coupling (15). When the avidity for distal tubular sodium-cation exchange is increased at a time when the distal delivery of sodium is adequate, then hydrogen ion secretion is greatly facilitated (2). This formulation appears to hold for explaining the renal mechanisms acting to increase acid excretion both

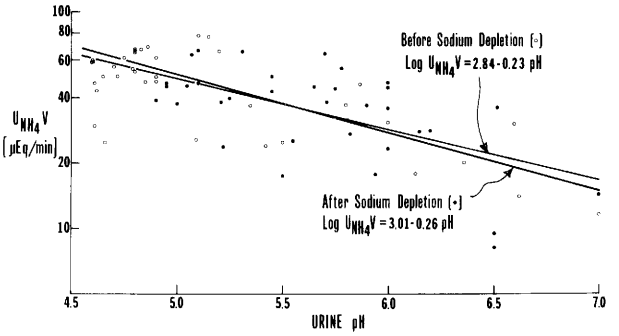

FIG. 2. Plot of $U_{NH4}V$ versus urine pH before and after sodium depletion. The open circles denote values prior to and the closed circles denote those after sodium depletion. The regression lines and their equations are shown for each group. (See text for explanation.)

during mineral acid loading and during the development of metabolic alkalosis (1).

The effect of sodium restriction on sodium reabsorption in various nephronal sites is not well defined. We have no direct data regarding distal sodium delivery in our patients. There is experimental micropuncture evidence (16) that distal sodium delivery is reduced during mild sodium depletion. In addition, the data of Stein *et al.* (17) demonstrating that salt restriction limits Tc H_2O formation in normal subjects are consistent with decreased distal delivery of sodium. On the other hand, recent studies (18) have demonstrated that the collecting duct is primarily responsible for the increased sodium reabsorption during moderate extracellular fluid volume depletion.

One might expect that urine pH might be higher and acid excretion lower if the distal delivery of sodium is limited. This might be the case, at least in association with some types of disordered renal acidification. For example, Buckalew *et al.* (19) reported that the urine pH-lowering capacity of a patient with the incomplete syndrome of distal renal tubular acidosis was limited further by dietary sodium restriction and Better *et al.* (20) as well as ourselves (21) have suggested that the abnormal renal acidification associated with alcoholic liver disease is related at least in part to the concomitant sodium-retaining state. Furthermore, enhancement of distal sodium delivery by diuretic administration has ameliorated the acidification impairment in some patients with distal RTA (22).

There has been very little systematic evaluation of the influence of sodium depletion on the renal response to acid loading. Our findings are similar to those of Clarke *et al.* (3) in that these authors noted a lower ammonium excretion (without a change in urine pH) when NH_4Cl acid loading followed sodium depletion. Both of the patients studied by Ryberg during a 10-day acid-loading test excreted urine containing less acid and less ammonium during periods of low sodium excretion (23). The ingestion of 4% NaCl was promptly followed by enhancement of ammonium excretion and a lowering of urine pH. Sodium balance data was not provided but one might suppose that the sustained NH_4Cl loading had resulted in a substantial negative sodium balance (23). After acid loading sodium-depleted subjects for 2–3 days, Schwartz *et al.* (4) demonstrated a relative enhancement of ammonium excretion in comparison to that reported by other investigators in sodium-replete volunteers. Their subjects, however, were not studied prior to sodium depletion and it is conceivable that their ammonium excretion was increased by some other variable or that distal sodium delivery was facilitated at the time of acid loading.

The findings in the present study are consistent with reports stressing the importance of distal tubular sodium reabsorption in renal acid handling. Although we have no direct data in our patients relevant to the distal delivery of sodium, it is tempting to speculate that in our sodium-depleted subjects maximal distal hydrogen ion secretion was limited by decreased distal delivery of sodium despite presumed increase in sodium avidity at the same nephronal site.

Summary. The effect of sodium depletion on urinary acidification was investigated in five healthy male volunteers. The response to short-duration NH_4Cl loading was assessed both before and during sodium depletion induced by low salt diet plus a single dose of furosemide. The minimal urine pH was higher ($P < 0.005$) and the maximal rate of ammonium excretion was lower ($P < 0.02$) following sodium depletion. These changes could not be explained by changes in acid–base status, serum potassium concentration, urine flow rate, or urinary phosphorus excretion. The findings support the concept that sodium delivery to distal exchange sites in concert with sodium reabsorptive avidity are major regulators of the renal response to acid loading. It is suggested that following sodium depletion a decreased distal delivery of sodium limited maximal hydrogen ion secretion despite the probable enhancement of the avidity for H^+-Na^+ exchange.

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