PROCEEDINGS OF THE SOCIETY FOR EXPERIMENTAL BIOLOGY AND MEDICINE 145, 737–742 (1974)

Factors Influencing Sodium Reabsorption in the Distal Nephron (37886)

THOMAS KAHN, EVEN ALFAGO, FREDERICK GISE,
DIMITRIOS A. PARDALIS, MARVIN F. LEVITT,
AND MARVIN H. GOLDSTEIN
(Introduced by M. Mendlowitz)

Division of Renal Diseases, Department of Medicine, Mount Sinai School of Medicine of the City University of New York, New York, New York 10029

The factors which influence sodium reabsorption in the nephron beyond the proximal tubule (distal tubule) remain unclear. Utilizing free water clearance (C_{H_2O}) as an index of distal-tubule sodium reabsorption, we have suggested that the development of a plateau in C_{H_2O} during hypotonic saline loading indicates that volume expansion limits the capacity of the distal nephron to reabsorb sodium (1). A maximal level of C_{H_2O} was subsequently observed during hypertonic saline loading in man with diabetes insipidus (2). Other workers, however, have repeatedly been unable to demonstrate any limit in C_{H_2O} in the dog (3–5) and have suggested that saline loading does not alter the characteristics of sodium reabsorption in the distal nephron (5). Moreover, a clear effect of volume expansion on the capacity of the loop of Henle to reabsorb sodium has not been demonstrated by microperfusion studies (6).

Previous clearance studies in the dog (7, 8) and recent micropuncture experiments in rats with congenital diabetes insipidus (9) have suggested that alterations in the passive back diffusion of water, even in the absence of antidiuretic hormone (ADH), may make the interpretation of C_{H_2O} as a measure of distal sodium reabsorption less useful. The present experiments were therefore performed in order to reevaluate whether a maximal level of C_{H_2O} is present in the dog and does reflect a limited capacity of the distal nephron to reabsorb sodium, and to permit further insight into other factors which may influence distal sodium reabsorption. Studies were performed with sodium sulfate in order to dissociate sodium chloride supply to the distal nephron from distal tubular sodium supply, solute load, and urine flow rate.

Methods. Experiments were performed on mongrel dogs weighing 20–29 kg. The preparation of the dogs was essentially as described in a previous report (1). Maximal hydration was achieved by the infusion of 2.5% glucose in water at 8–20 ml/min until urine osmolality was less than 100 mOsm/kg water.

In 25 studies, 0.7–1.0% sodium sulfate was administered initially at a rate of 15 ml/min and subsequently increased to exceed urine flow rate by 10–15 ml/min. In 13 of the 25 studies, 0.3% sodium chloride was administered with 0.7% sodium sulfate. In 14 studies, a Blalock clamp was initially loosely positioned around one renal artery. During the sodium sulfate infusion, after control urine collections had been obtained from both kidneys, the clamp was tightened around the renal artery to produce an ipsilateral decrease in urine flow rate. Subsequently, the rate of the sodium sulfate infusion was increased as described above.

Blood and urine were analyzed for osmolality, sodium, potassium, chloride, and inulin by standard methods (1).

Results. The infusion of sodium sulfate

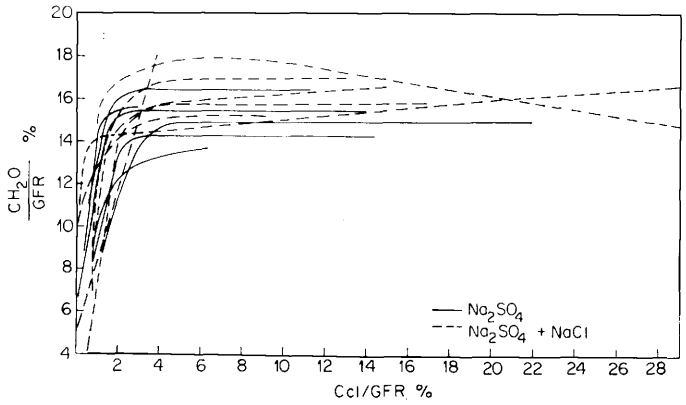

FIG. 1. Relationship between the percent of the GFR excreted as C_{H_2O} (C_{H_2O}/GFR × 100) and the percent of the filtered chloride load excreted (C_{Cl}/GFR × 100) in a single kidney in studies in which fractional chloride excretion exceeded 2% during the administration of hypotonic sodium sulfate (—) and hypotonic sodium sulfate plus sodium chloride (- - -).

alone produced results which were indistinguishable from those in which sodium chloride was administered with sodium sulfate (Figs. 1, 2, and 4). Therefore, the results of all studies have been treated together. Glomerular filtration rate (GFR) and plasma sodium remained stable throughout these studies whereas plasma chloride fell progressively. Since sulfate is only minimally reabsorbable (10), a consistent natriuresis and solute diuresis were produced in each study.

In 16 studies in which GFR in a single kidney averaged 33 ml/min, the fraction of the filtered chloride load excreted in the urine (C_{Cl}/GFR) exceeded 2%. In these studies, as C_{osm} and V increased, C_{H_2O} initially rose and then stabilized at maximal levels (Figs. 1 and 3). Maximal C_{H_2O}/GFR ranged from 13 to 18%, averaging 15% (Figs. 1 and 3). C_{Cl}/GFR was less than 2% prior to the attainment of the maximal levels of C_{H_2O}, and subsequently fractional chloride excretion increased with peak values averaging 12% and reaching levels as high as 29% (Figs. 1 and 3).

In nine experiments in which GFR averaged 31 ml/min, the fraction of the filtered chloride excreted remained below 2%. C_{osm} and V increased progressively in these studies to levels comparable to those achieved in the experiments in which fractional chloride excretion exceeded 2% (Fig. 2). When C_{H_2O} formation was compared at the same level of C_{osm} (or V) (Fig. 2), C_{H_2O} was clearly higher in the studies in

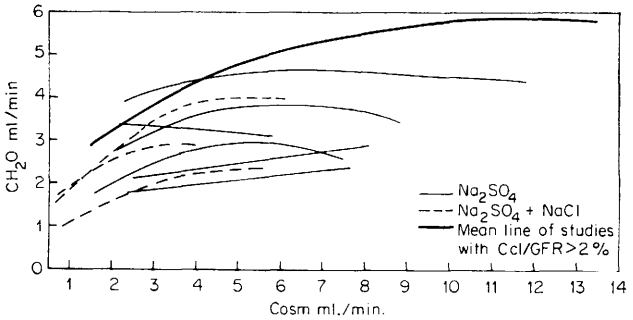

FIG. 2. Relationship between C_{H_2O} and C_{osm} in a single kidney in those studies in which fractional chloride excretion remained below 2% during the administration of hypotonic sodium sulfate (—) and hypotonic sodium sulfate plus sodium chloride (- - -). The mean line for studies in which fractional chloride excretion exceeded 2% is given for comparison.

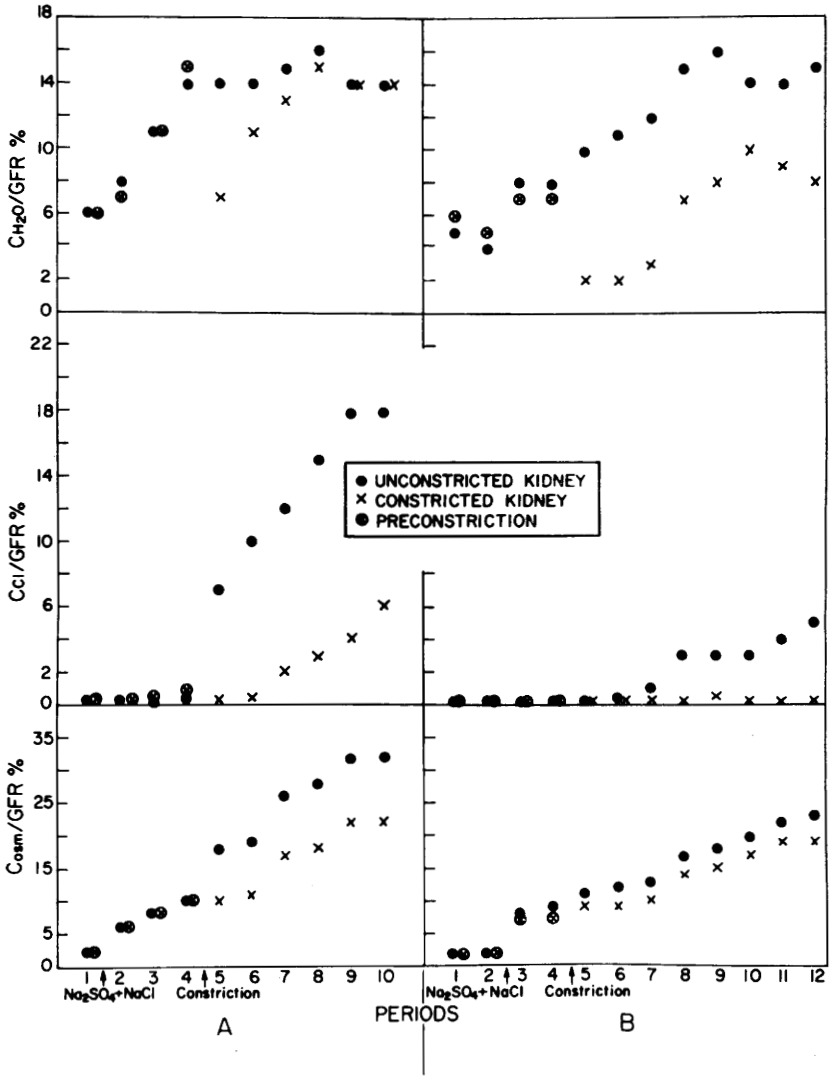

FIG. 3. Two representative experiments depicting the effects of unilateral renal artery constriction on C_{H_2O}/GFR, C_{Cl}/GFR, and C_{osm}/GFR in the constricted (×) and contralateral kidney (●) during successive urine-collection periods during the administration of hypotonic sodium sulfate plus sodium chloride. C_{Cl}/GFR eventually exceeded 2% in the constricted kidney in A and remained below 2% in B.

which fractional chloride excretion exceeded 2%. The highest level of C_{H_2O}/GFR attained in the studies in which fractional chloride excretion remained below 2% ranged from 6 to 12%.

Constriction of one renal artery resulted in a fall in GFR in the ipsilateral kidney to 53–89% of the GFR in the contralateral kidney. Urine flow rate and C_{osm} fell initially on the constricted side but rose as the sodium sulfate infusion produced a progressive diuresis from both kidneys (Fig. 3). C_{H_2O} formation following renal artery constriction again appeared to be related to chloride excretion. In six studies, C_{Cl}/GFR eventually exceeded 2%, and in eight studies C_{Cl}/GFR remained less than 2% on the constricted side. In the former studies, C_{H_2O}/GFR on the constricted side reached levels comparable to those achieved

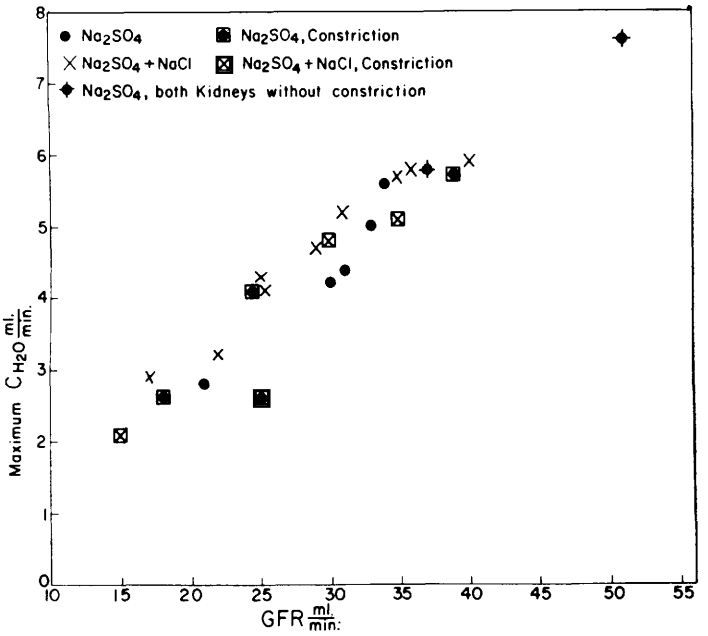

FIG. 4. Each point represents the average maximal C_{H_2O} in ml/min in a single kidney from studies in which C_{Cl}/GFR × 100 exceeded 2% plotted against the GFR of that kidney. Included in the figure are studies in which hypotonic sodium sulfate alone (●) and hypotonic sodium sulfate plus sodium chloride (×) was administered. Kidney with renal artery constriction in which C_{Cl}/GFR exceeded 2% are depicted (⊡, ⊠). Both kidneys are represented from one study in which a stable spontaneous difference in GFR was noted (+).

in the contralateral kidney (Fig. 3A). At any point in time, however, C_{Cl}/GFR and C_{osm}/GFR were always lower in the constricted side than in the contralateral kidney (Fig. 3A). In the studies in which C_{Cl}/GFR remained less than 2%, C_{H_2O}/GFR was lower on the constricted side than on the contralateral side and remained lower as solute excretion progressively increased (Fig. 3B).

The maximal level of C_{H_2O} per kidney in all studies in which fractional chloride excretion exceeded 2% was plotted versus the GFR in that kidney (Fig. 4). Included in this plot are data from renal-artery-constriction experiments and in one instance from both kidneys of one dog with a spontaneous stable difference in GFR. A direct linear relationship between maximal C_{H_2O} and GFR was apparent (Fig. 4). Maximal levels of C_{H_2O} ranged from 2.2 to 7.6 ml/min and GFR from 15 to 51 ml/min. Maximal C_{H_2O}/GFR, however, varied only from 13 to 18%, averaging 15% (Figs. 1 and 3).

Discussion. It is generally accepted that despite either increasing (9) or decreasing (7, 8, 11) back diffusion of water in the distal nephron during a solute diuresis, an increase in C_{H_2O} is primarily consequent to increasing sodium reabsorption in the distal nephron in excess of the back diffusion of water (1–5). The finding in the present studies that C_{H_2O} reached a plateau suggests either that distal sodium chloride reabsorption did not increase appreciably or that the back diffusion of water increased so markedly that it completely masked further sodium chloride reabsorption. The latter possibility seems particularly unlikely to occur during solute diuresis in dogs in which the passive back diffusion of water has previously been shown to decrease (7, 8, 11) and the medullary tonicity, which is the stimulus to water back diffusion, either fails to increase (3) or actually decreases (12). Moreover, the temporal relationship between the attainment of a

plateau in C_{H_2O} and the onset of a progressive chloruresis in the present studies would not be explained by increased back diffusion of water. This relationship suggests that the plateau in C_{H_2O} does reflect a limit in distal-tubule sodium reabsorption. In the studies with minimal chloride excretion, and presumably less chloride supply to the distal nephron, C_{H_2O} did not achieve levels comparable to that noted in the studies in which fractional chloride excretion averaged 12% (Fig. 2). When chloride excretion was severely restricted by renal-artery constriction, presumably associated with a marked reduction in distal chloride supply, absolute and fractional C_{H_2O} were reduced and did not attain comparable levels to that noted in the contralateral kidney (Fig. 3B). When fractional chloride excretion from the constricted kidney reached appreciable levels, C_{H_2O}/GFR reached comparable levels to that noted in the control kidney (Fig. 3A). Thus, the level of C_{H_2O} generated appears to relate primarily to the supply of sodium chloride reaching the distal nephron. Although some quantity of water may back diffuse during a solute diuresis, the maximal level of C_{H_2O} attained in the present studies in which chloride excretion exceeds 2% does appear to represent an index of a maximal capacity of the distal nephron to reabsorb sodium chloride.

Maximal C_{H_2O}/GFR during sodium sulfate loading averaged 15%, a value similar to that obtained in previous studies with saline loading in dog (1). Since higher levels of C_{H_2O}/GFR were obtained in dogs with mannitol loading (1) and in man with the administration of acetazolamide alone (2), the maximal level of C_{H_2O} does not appear to be an intrinsic limit in distal sodium chloride reabsorption. Moreover, since C_{H_2O} increased progressively when sufficient sodium chloride reached the distal nephron during mannitol loading (1), the limit in C_{H_2O} in the present studies cannot be attributed to sodium sulfate acting as a nonspecific solute load. Therefore, the limit in C_{H_2O} appears to reflect an alteration in the capacity of the distal nephron to reabsorb sodium chloride consequent to volume expansion with a sodium salt.

The present studies permit an analysis of factors which may influence the rate of distal sodium reabsorption. During sodium sulfate loading, plasma chloride concentration and, therefore, tubular chloride concentration fell markedly. The finding that maximal C_{H_2O} was similar in the sulfate and saline studies suggests that lowering distal tubular chloride concentration does not impair the capacity of the distal tubule to reabsorb sodium chloride. The level of maximal C_{H_2O} did not correlate with the rate of solute excretion (Fig. 2) or urine flow rate, suggesting that these factors do not exert the major influence on the capacity of the distal nephron to reabsorb sodium chloride.

On the other hand, the maximal level of C_{H_2O} in the present experiments did correlate with GFR (Fig. 4), an observation made previously by other workers (2). Spontaneous differences in GFR in mongrel dogs of similar weight and differences in GFR induced by renal-artery constriction in the present studies are probably consequent to the various physiologic factors which determine GFR per nephron, rather than differences in the number of functioning nephrons per kidney (13–15). Thus, the relationship between GFR and the maximal level of C_{H_2O} suggests that those factors which determine the filtration rate may also influence the maximum capacity of the distal nephron to reabsorb sodium chloride under conditions of volume expansion. Although the influence of these factors may be most easily demonstrable at maximal rates of distal sodium reabsorption, it seems likely that these factors influence the rate of distal sodium reabsorption prior to the attainment of a maximal rate. Many workers have demonstrated that a reduction in GFR is associated with a decrease in sodium reabsorption in the proximal nephron, thus permitting sodium chloride to reach distal tubular sites. A decrease in the capacity of the distal nephron to reabsorb sodium under these conditions may help to maintain the excretion of

sodium chloride in a kidney with a diminished filtration rate.

Summary. In 16 of 25 studies during hypotonic sodium sulfate infusion, the development of a chloruresis was temporally associated with a maximal level of C_{H_2O}. In nine studies, no chloride diuresis developed, and C_{H_2O} remained lower than in the former group despite the attainment of similar levels of solute excretion and urine flow rate. With renal-artery constriction, a close relationship was also demonstrated between the level of C_{H_2O} and the supply of sodium chloride to the distal nephron. The level of C_{H_2O} attained did not relate to levels of solute excretion, urine, flow rate, or intratubular chloride concentration. However, a direct linear relationship was observed between the maximal level of C_{H_2O} and GFR over a range of GFR from 15 to 51 ml/min. These findings support the view that a maximal level of C_{H_2O} represents a limit in sodium chloride reabsorption in the distal nephron and suggest that the capacity of the distal nephron to reabsorb sodium chloride is influenced by those factors determining the glomerular filtration rate.

We thank Mrs. Sarah Bogert for her technical assistance. This work was supported by Public Health Service Training Grant HE 5-5505 and a grant from the New York Kidney Foundation.

1. Stein, R. M., Abramson, R. G., Kahn, T., and Levitt, M. F., J. Clin. Invest. **46**, 1205 (1967).

2. Buckalew, V. M., Walker, B. R., Puschett, J. B., and Goldberg, M., J. Clin. Invest. **49**, 2336 (1970).

3. Eknoyan, G., Suki, W. N., Rector, F. C., Jr., and Seldin, D. W., J. Clin. Invest. **46**, 1178 (1967).

4. Rosin, J. M., Katz, M. A., Rector, F. C., Jr., and Seldin, D. W., Am. J. Physiol. **219**, 1731 (1970).

5. Barton, L. J., Lackner, L. H., Rector, F. C., Jr., and Seldin, D. W., Kidney Int. **1**, 19 (1972).

6. Morgan, T., and Berliner, R. W., Nephron **6**, 388 (1969).

7. Orloff, J., Wagner, H. N., and Davidson, D. G., J. Clin. Invest. **37**, 458 (1958).

8. Aukland, K., and Kjekshus, J., Amer. J. Physiol. **210**, 971 (1966).

9. Jamison, R. L., and Lacey, F. B., Amer. J. Physiol. **210**, 971 (1966).

10. Lotspeich, W. D., Amer. J. Physiol. **151**, 311 (1947).

11. Earley, L. E., Kahn, M., and Orloff, J., J. Clin. Invest. **40**, 857 (1961).

12. Boonjarern, S., Stein, J., Hsueh, W., Cohen, S., Yashon, D., and Ferris, T., Clin. Res. **20**, 761 (1972).

13. Thompson, D. D., Barrett, M. J., and Pitts, R. F., Amer. J. Physiol. **167**, 546 (1951).

14. Orlowski, T., and Bricker, N. S., Acta Med. Polona **3**, 247 (1964).

15. Liebau, G., Levine, D. Z., and Thurau, K., Pflugers Arch. Ges. Physiol. **304**, 57 (1968).

Received Oct. 15, 1973. P.S.E.B.M., 1974, Vol. 145.