Effect of Reduced Renal Mass on Ammonium Handling and Net Acid Formation by the Superficial and Juxtamedullary Nephron of the Rat

EVIDENCE FOR IMPAIRED REENTRAPMENT RATHER THAN DECREASED PRODUCTION OF AMMONIUM IN THE ACIDOSIS OF UREMIA

JOHN BUERKERT, DANIEL MARTIN, DAVID TRIGG, and ERIC SIMON, *Renal Physiology Laboratory of The Jewish Hospital of St. Louis, Washington University School of Medicine, St. Louis, Missouri 63110*

ABSTRACT Papillary and surface micropuncture were used to study the handling of ammonium and the formation of net acid by surface nephrons, deep nephrons, and the terminal segment of collecting duct (CD) after renal mass was reduced by two-thirds. Net acid excretion by the remnant kidney (RK) was significantly reduced, averaging 794±81 neq/min (SE) compared with 1,220±105 neq/min after sham operation ($P < 0.001$), due to a decrease in ammonium excretion (494±54 vs. 871±79 nmol/min in controls, $P < 0.001$). Urinary pH and titratable acid excretion were not different in the two groups of animals. After RK formation, ammonium delivery to the end of the proximal tubule increased nearly threefold and averaged 66.2±5.6 compared with 18.4±2.9 pmol/min in controls, ($P < 0.001$). This greater delivery of ammonium was primarily due to renal tubule entry rather than to changes in the filtered load and was only partially related to the differences in flow rate. Ammonium processing by deep nephrons was profoundly affected by a reduction in renal mass. Although absolute delivery of ammonium was greater to the bend of Henle's loop (BHL), the difference could be accounted for on the basis of an increase in nephron size. Thus, fractional delivery ($FD_{NH_4^+}$) to this site was not different for the two groups of animals, averaging 1,567±180% in controls and 1,400±181% in the group with the RK. Hydrogen secretion in the proximal segments of deep and surface nephrons did not increase in proportion to the decrease in renal mass and as a consequence bicarbonate delivery to the end of the proximal tubule of surface nephrons and to the BHL of deep nephrons was increased.

When renal mass was reduced $FD_{NH_4^+}$ to the base of the terminal CD doubled but did not change by the tip. In both groups $FD_{NH_4^+}$ to the base of the CD was greater than to the end of the distal tubule. However, the increase was the same. On the other hand, the increase in the net acid index between the end of the distal tubule and the base of the CD was profoundly greater in rats with an RK. This difference was primarily due to bicarbonate reabsorption rather than enhanced ammonium reentry. Indeed, >400% of the fractional ammonium delivered to the end of the proximal tubule was lost from the tubule fluid. The data suggest that the decrease in acid excretion by the RK is due to two factors. First, hydrogen secretion in the proximal segments of both nephron populations fails to increase in the proportion to the reduction in renal mass. Second, a reduced reentrapment of ammonia, rather than its impaired production, causes ammonium excretion to decrease.

INTRODUCTION

One early consequence of a reduction in renal mass is the development of a metabolic acidosis, a reflection of the inability of the residual nephrons to excrete the hydrogen ion generated through the metabolic degradation of dietary protein (1–4). At least two different mechanisms are responsible for this altered acid ex-

Portions of this work were presented to the 39th Annual Meeting of the American Society of Clinical Research, Wash. DC, 10 May 1982.

Received for publication 1 November 1982 and in revised form 23 February 1983.

cretion. First, it appears that secretion of hydrogen ion in the proximal tubule, as it relates to bicarbonate reabsorption, fails to keep pace with the increase in the filtered load of bicarbonate per nephron that occurs as renal function declines. This suggestion is supported by whole kidney clearance studies in humans (4, 5) and experimental animals (6–8) with reduced renal function, and appears to be a consequence of uremia per se, since the diseased kidney functioning in the presence of the contralateral control kidney (CK)[1] does not "waste" bicarbonate (6, 9). The micropuncture studies of Lubowitz et al. (6) have implicated the proximal tubule in this altered reabsorption of bicarbonate. However, beyond these studies, the handling of bicarbonate by the various nephron segments of the chronically diseased kidney have not been assessed. No information is available concerning the role of the proximal segments of deep nephrons in bicarbonate reclamation in the setting of reduced renal mass.

The second mechanism operative in the metabolic acidosis of uremia is the reduced excretion of ammonium (4, 10–13). In humans (4, 11, 12) and in animals (14–17) with reduced renal mass, ammonium excretion does not increase appropriately when an acid load is administered chronically or when the substrate for ammonia production, glutamine, is given (13). Although ammonium excretion seems to bear a proportional relationship to the degree of reduction in nephron mass (11, 12), there is evidence that an adaptive increase occurs. Studies of unilateral renal disease have shown that the excretion of ammonium by the diseased kidney is similar to that of the contralateral kidney when factored by glomerular filtration rate (GFR) (9, 16). However, when the normal functioning kidney is removed (9, 16), ammonium excretion/100 ml of GFR by the remnant kidney (RK) increases significantly. Similarly, several investigators have shown that ammonium excretion per nephron is increased in this setting (15–17). Finkelstein and Hayslett (15) have reported a more profound decrease in ammonium and titratable acid excretion in rats after papillectomy than after partial renal ablation. These data suggest that papillary structures play an important role in the excretion of buffered acids. Except for these observations no studies have been performed that are directed toward the specific nephron site or sites where altered ammonium handling occurs after renal mass is reduced.

Recently, we (18, 19) and others (20) have provided evidence, using micropuncture techniques, suggesting that ammonium production occurs primarily in the proximal segments of surface and deep nephrons. That ammonium leaves surface nephrons between the end of the proximal and distal tubule to become reentrapped along the medullary collecting duct (CD) (18–21). Thus, it would appear that the process of acidification involves two steps, either of which might be affected by a reduction in renal mass. This study was therefore designed to characterize the relative contribution of deep and surface nephrons of the RK to the process of net acid formation and ammonium production and to determine whether the reduction in ammonium excretion seen after renal mass is reduced is due to decreased production in the proximal segments of these two nephron populations or to a failure to reentrap ammonia along the terminal nephron and the CD.

METHODS

Approximately 14 d before study, 21 Munich-Wistar rats weighing 60–70 g were lightly anesthetized with ether. The left kidney was exposed through a midline abdominal incision and the renal pedicle was carefully dissected free of the surrounding adipose tissue. The lower pole, upper pole, and posterior surface of the kidney were infarcted by ligating the secondary and tertiary branches of the renal artery with 6-0 silk suture. 1 wk later the rats were again anesthetized with ether and a right nephrectomy was performed. Approximately 5–7 d later, the rats were prepared for study. In 11 rats, the renal papilla was exposed and prepared for micropuncture as previously described (22). In the remaining 10 rats, a piece of polyethylene tubing (PE No. 50) was placed in the bladder and standard clearance studies were performed. 25 rats underwent sham operation ~14 d before study, the time between renal infarction and study in the group with the RK. In 10 of the sham-operated animals, only clearance studies were performed; in the remaining 15, the left kidney was prepared for papillary micropuncture (22). In all rats, food but not water was withheld for ~12 h before study.

On the day of micropuncture the rats were anesthetized with Inactin (Promonta, Hamburg, West Germany). Immediately after the abdomen was opened but before the kidney was prepared for study, all rats were given an inulin prime in 1 ml of 0.9% NaCl. This was followed by an infusion of normal saline containing inulin in sufficient amounts to maintain the plasma concentration >50 mg/dl. The infusion rate was calculated at 35 μl/min per 100 g body wt. This rate of fluid replacement resulted in a stable hematocrit. The mean value was 42.2±3.3% in the control and 39.6±0.6% in the remnant group at the beginning of the study, and 41.4±0.8 and 39.9±0.6%, respectively, at the end of the experiment. These values were slightly lower ($P < 0.01$) than those obtained before the institution of fluid replacement (44.9±0.8% in controls and 41.3±0.6% in the RK group). In all studies an equilibration period of 45–60 min was allowed.

Collection from the papillary structures was instituted 20–30 min following removal of the ureteral pelvis. In each group, two to five loops were identified visually and timed tubule fluid samples were collected. Immediately after the collection of each timed sample a microquinhydrone pipet was introduced into the tubule site of the previous puncture.

[1] *Abbreviations used in this paper:* BHL, bend of Henle's loop; CD, collecting duct; CK, control kidney; GFR, glomerular filtration rate; RK, remnant kidney; SNGFR, single nephron GFR;($T_{NH_4^+}$), renal tubular secretion of ammonium.

In most cases, fluid spontaneously entered the pipet and when a sufficient amount was collected to cover the end of the electrode, the pipet was removed and the tip immediately sealed with egg albumin. In all cases the sample size was <10 nl, as judged visually. Samples were then obtained from CD at sites as close to the base of the papilla (CD_{prox}) as possible and from points along the same CD near the tip (CD_{tip}). The distance between the two collecting duct sites was estimated with an eyepiece micrometer. In almost all cases, fluid samples were obtained from at least two different CD. In situ pH was then determined at the sites of previous collection using double-barreled pipettes consisting of a glass membrane pH microelectrode and a reference electrode.

Either before or after collections from papillary structures were obtained, the cortical surface was illuminated with a fiberoptics light guide attached to a quartz rod. The end accessible portion of proximal and distal tubules was identified by a small amount of 5% lissamine green (0.02–0.05 ml) injected intravenously. After the dye disappeared from the surface of the kidney timed tubule fluid collections were obtained, in situ pH measurements were made, and fluid was collected into a microquinhydrone electrode.

The methods for the measurements of volume and the tubule fluid concentration of sodium, potassium, osmolality, and inulin are reported elsewhere (22). The concentration of ammonium and titratable acid in the tubule fluid sample was determined with a microtitrating apparatus that has been described in prior studies (18, 19, 23). The bicarbonate concentration of tubule fluid was calculated from in vitro pH measurements determined with the microquinhydrone electrodes and a modification of the Henderson-Hasselbalch equation (19). The precision and accuracy of these methods have been detailed in previous studies (18, 19). The double-barreled pH microelectrodes for both titration of tubule fluid samples and in situ pH measurements were constructed with pH-sensitive glass as described by Pucacco and Carter (24). The pH-sensitive side of the microelectrode was filled with 1 mM magnesium acetate saturated with silver chloride. The reference side contained a solution of 2.5 M KCl and 0.5 M KNO_3. The potential difference between the two sides of the microelectrode was sensed by an electrometer (model 602C, Keithley Instruments, Cleveland, OH) to which they were connected by silver chloride wires. In all cases the reported values were obtained after the measurement had remained stable for 45–60 s. These electrodes were checked with buffers of known pH before and after in situ pH measurements had been made. The slope of the pH electrodes used in the study averaged 58.1±0.07 mV/pH unit.

Blood samples and blood pressure measurements were obtained at hourly intervals or immediately after the collection of tubule fluid. Body temperature was determined with a rectal thermometer and maintained at 36°–38°C. In sham-operated rats, one or two timed collections of urine were made in preweighed test tubes and the volume was determined gravimetrically. The methods for determination of inulin, sodium, potassium, osmolality, blood urea nitrogen (BUN) in plasma and urine have been described previously (22). Arterial pH and P_{CO_2} was measured hourly with a blood gas microsystem (BMS-3, Radiometer America, Inc., Westlake OH). Urine and plasma ammonium concentration was determined with an ion specific electrode and a flow through cell (Orion Research, Inc., Cambridge, MA) as previously described (18).

Tubule fluid-to-plasma inulin ratios (TF/P_{In}) permitted the calculation of fractional fluid reabsorption before the site of micropuncture. Single nephron glomerular filtration rate (SNGFR) and fractional delivery of fluid (FD), and ions to the site of micropuncture were determined using standard formulas.

At proximal sites, renal tubule secretion of ammonium (T_{NH^+}) was calculated according to the following formula: $T_{NH^+} = (TF_{NH^+}V_{tf}) - (FL_{NH^+})$, where $TF_{NH^+}V_{tf}$ is the absolute delivery of NH^+ to each site of micropuncture and FL_{NH^+} is the single nephron filtered load of this cation (SNGFR $\times$ P_{NH^+}). Entry of ammonium between the end of the proximal and distal tubule [$(T_{NH^+})_{Distal}$] was calculated as the difference in the ammonium delivered to these sites: $(T_{NH^+})_{Distal} = (TF_{NH^+}V_{tf})_{Distal} - (TF_{NH^+}V_{tf})_{Proximal}$. Mean values for absolute delivery of ammonium delivered to the end of the proximal and distal tubule for each rat were used in these latter calculations.

The net acid content of tubule fluid samples was calculated as follows: $TF_{NA} = (TF_{TA}) + (TF_{NH_4^+}) - (TF_{HCO_3^-})$, where TF is the tubule fluid concentration of net acid (NA), titratable acid (TA), ammonium (NH_4^+), and bicarbonate HCO_3^-). The bicarbonate content of CD fluid was not determined. Because of the relatively low pH it seemed justified to assume that $TF_{HCO_3^-}$ did not contribute significantly to the overall measurements. Thus, $TF_{NA} = TF_{TA} + TF_{NH_4^+}$. The rate of hydrogen secretion before the site of micropuncture was assumed to be $TF_{H^+}V_{tf} = TF_{TA}V_{tf} + TF_{NH_4^+}V_{tf} + T_{HCO_3^-}$, where TF_xV_{tf} is the absolute delivery of TA and NH_4^+ to the site of micropuncture and $T_{HCO_3^-}$ is the absolute rate of bicarbonate reabsorption before this site. The index for titratable acid, ammonium, and net acid was calculated according to the following formula: $TF_x/(TF/P_{In})$, where x is the TA, NH_4^+, and NA content of the sample.

The mean values for individual micropuncture samples obtained at similar anatomical sites for each rat were used for statistical analysis. Mean differences in whole kidney and superficial nephron function were examined by the Student's *t* test for unpaired data when comparing the two groups of animals studied and for paired data when comparing superficial and deep nephron functions in the same animal or when comparing proximal and tip measurements along the same CD. When indicated, samples were compared statistically after logarithmic transformation (25).

RESULTS

Table I summarizes the base-line information obtained in the two groups of rats. Micropuncture and clearance studies were performed 11.4±2.8 d (SD) after sham operation and 12.9±1.9 d after left renal infarction. There was no statistical difference in the body weights of the two groups of animals at the time of the study. The mean weight of the RK was slightly more than a third of the values obtained for the total renal mass of the sham-operated group. This reduction in renal mass was reflected in a significant rise in BUN (from 20.7±0.9 to 59.4±6.1 mg/dl [SE], $P < 0.001$). At the time of study, rats with an RK exhibited a mild metabolic acidosis; both the arterial pH and bicarbonate concentration were significantly lower in this group than in the sham-operated group. Plasma ammonium concentrations were not different in the two groups of animals.

As reported previously (22), the general appearance of the viable portion of the kidney in the two groups

TABLE I

Weight, Blood Pressure, and Plasma Parameters in Sham-operated Rats and Rats with an RK

	Body weight	Kidney weight	Blood pressure	Plasma levels				
				BUN	pH	P_{CO_2}	HCO_3^-	NH_4^+
	g	*mg*	*mmHg*	*mg/dl*		*mmHg*		*mM*
Control group (n = 25)	96.7±1.9	866±20	113±2	20.7±0.9	7.37±0.01	36.2±0.7	20.3±0.4	0.11±0.01
Remnant group (n = 21)	95.4±2.9	307±21	122±3	59.4±6.1	7.31±0.02	35.9±1.0	17.9±0.5	0.12±0.01
P	NS	<0.001	NS	<0.001	<0.005	NS	<0.005	NS

Values are the mean±SE. n, number of rats studied in each group. P, level of significance when the two groups are compared. Kidney weight refers to the weight of the viable portion of the RK and the weight of the right and left kidney of the control group.

of animals was the same. In the remnant group, the area of functioning renal tissue formed a central band with scar tissue present on the posterior surface and both superior and inferior poles. Microscopically, renal tubules on the surface of the RK appeared larger and more dilated than those of the CK. The appearance of the papilla available for study was not different in the two groups of rats. The mean length averaged 1.1±0.1 mm for the RK compared with 1.2±0.1 mm for the CK.

Clearance studies

The mean values for whole kidney function obtained in 10 sham-operated rats and in 10 rats with an RK are presented in Table II. The GFR of the RK averaged 523±62 μl/min, a third of the mean obtained in the control group, 1,366±114 μl/min. However, because of the threefold increase in fractional fluid excretion by the RK (from 0.365±0.043 to 1.05±0.48%, $P < 0.01$), urine flow (V) did not differ statistically in the two groups. Mean values for urine pH obtained in the control and remnant group were virtually the same. However, the total buffered acid excreted, that is, net acid excretion, was reduced in the group with the RK (794±81 vs. 1,220±105 neq/min in controls, $P < 0.001$). This decrease was primarily due to a reduction in the excretion of ammonium, which averaged 494±54 nmol/min, in contrast to 871±79 nmol/min for the control group ($P < 0.001$). Nevertheless, ammonium excretion by the RK rose relative to the filtered load. Fractional excretion averaged 1,154±142% in contrast to 803±107% for controls ($P < 0.05$) indicating that adaptation in ammonium production by the remaining nephrons occurred in this setting.

Nephron function

Surface nephrons. SNGFR measurements obtained near the end of the proximal tubule of the RK averaged 27.8±2.6 nl/min, which was not different from the mean obtained at end distal sites (Table III). However, both values were significantly greater than measurements obtained in the sham-operated group, which averaged 14.7±0.9 and 13.3±1.6 nl/min, respectively ($P < 0.005$). As in previous studies (22), delivery of fluid to the end of the proximal and distal tubule of the RK was significantly greater than delivery to these sites in the CK. In absolute terms, fluid delivery to the end of the proximal tubule averaged 17.3±1.9 nl/min in the remnant group in contrast to only 7.5±0.5 nl/

TABLE II

Whole Kidney Function in the Two Groups of Rats

	V	GFR	V/GFR	U_{pH}	$FE_{NH_4^+}$	$U_{NH_4^+}V$	$U_{TA}V$	$U_{NA}V$
	μl/min		*%*		*%*	*nmol/min*		*neq/min*
Control group n = 10	4.88±0.66	1,366±114	0.365±0.043	5.51±0.21	803±107	871±79	351±34	1,220±105
Remnant group n = 10	4.05±0.44	523±62	1.05±0.48	5.51±0.06	1,154±142	494±54	303±45	794±81
P	NS	<0.001	<0.01	NS	<0.05	<0.001	NS	<0.001

Values are mean±SE. V, rate of urine flow; V/GFR, fraction of filtered water excreted; U_{pH}, urine pH; $FE_{NH_4^+}$, fractional excretion of NH_4^+; U_xV, absolute titratable acid (TA), ammonium (NH_4^+), and net acid (NA) excretion.

TABLE III

Effect of a Reduction in Renal Mass on Acid Delivery to End Proximal and Distal Micropuncture Sites of Surface Nephrons

	SNGFR	V_{tf}	TF_{pH}	TF_{TA}	$TF_{TA}V_{tf}$	$TF_{NH_4^+}$	$TF_{NH_4^+}V_{tf}$	$T_{NH_4^+}$	$FD_{NH_4^+}$	$TF_{HCO_3^-}$	$TF_{HCO_3^-}V_{tf}$	$FD_{HCO_3^-}$
	nl/min			*meq/liter*	*peq/min*	*mM*	*pmol/min*		*%*	*mM*	*pmol/min*	*%*
Control group												
End proximal	14.7	7.5	6.92	1.01	7.09	2.51	18.4	16.6	1052	6.97	54.4	19.3
n = 14	±0.9	±0.5	±0.05	±0.11	±1.20	±0.18	±1.9	±1.9	±70	±0.76	±6.9	±2.2
End distal	13.3	2.11	6.70	2.22	5.42	5.56	10.7	−9.1	655	4.26	8.76	4.16
n = 10	±1.6	±0.34	±0.07	±0.39	±1.25	±0.68	±1.9	±2.3	±74	±1.12	±2.33	±1.29
P	NS	NS	<0.005	<0.01	NS	<0.005	<0.005	<0.001	<0.001	<0.05	<0.005	<0.005
Remnant group												
End proximal	27.8*	17.3‡	6.75§	1.89*	31.1	4.17‡	66.2‡	62.3‡	1823‡	7.79	134*	29.4§
n = 11	±2.6	±1.9	±0.09	±0.25	±5.0	±0.38	±5.6	±5.5	±194	±0.83	±20	±3.7
End distal	28.0*	7.03‡	6.61	4.84‖	31.8	5.02	29.3‡	−38.2‡	941	8.10§	58.8*	13.7§
n = 10	±3.5	±1.02	±0.12	±0.78	±7.6	±1.13	±5.5	±5.0	±174	±0.85	±12.5	±3.3
P	NS	NS	NS	<0.001	NS	NS	<0.001	<0.001	<0.001	NS	<0.005	<0.005

Values are the mean±SE. n, number of rats; TF_{pH}, in situ pH; TF, tubule fluid concentration of titratable acid (TA), ammonium (NH_4^+), and bicarbonate (HCO_3^-); TF_xV_{tf}, the absolute delivery of TA, NH_4^+, and HCO_3^- to the site of micropuncture; $T_{NH_4^+}$, was calculated at proximal sites as the difference between the absolute delivery and the filtered load of ammonium. At distal sites it was the difference in delivery to end proximal and distal micropuncture sites; FD_x, fractional delivery of HCO_3^- and NH_4^+ to micropuncture site. Values are significantly different from those obtained in controls:

* $P < 0.005$.

‡ $P < 0.001$.

§ $P < 0.05$.

‖ $P < 0.01$.

min in the sham-operated group. There was a similar increase in fractional fluid delivery to this site (from 51.6±2.4 to 61.2±2.4%, $P < 0.01$). Fluid delivery to end distal sites averaged 7.03±1.02 nl/min after renal mass was reduced, threefold greater than the mean of controls (2.11±0.34 nl/min, $P < 0.001$). Fractional delivery of fluid to the end of the distal tubule of the RK increased similarly from 16.1±1.8 to 27.0±4.0% ($P < 0.025$).

Acid measurements were made at 26 end proximal and 18 end distal micropuncture sites in the group with the RK. Similar measurements were made near the end of 33 proximal and 16 distal tubules of surface nephrons in the sham-operated group. Data obtained for both groups of animals are summarized in Table III. In situ pH measurements made near the end of the proximal tubule averaged 6.75±0.09 in the group with the RK. This value was slightly lower than the mean obtained at this site in the control group (6.92±0.05). This small increase in the free acid concentration was associated with a marked increase in the total buffered acid content of fluid delivered to this site. Tubule fluid titratable acid concentration was almost twofold greater in the RK group, averaging 1.89±0.25 meq/liter compared with 1.01±0.11 meq/liter for controls ($P < 0.005$). Absolute delivery of titratable acid to this site was nearly fivefold greater,

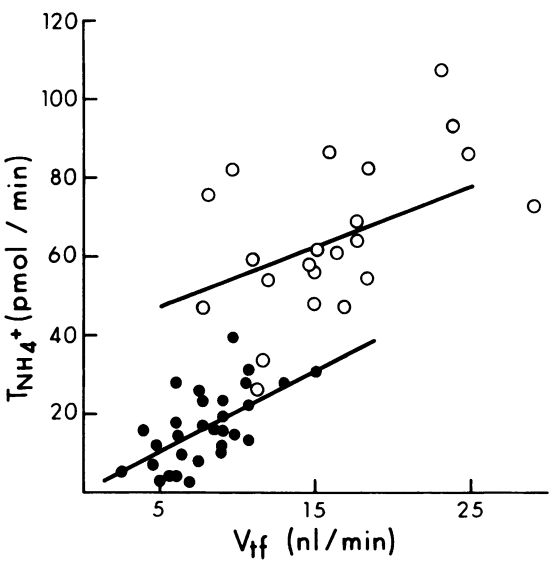

FIGURE 1 The relationship between entry of ammonia (T_{NH^+}) and tubule fluid flow rate (V_{tf}) in the control group (closed circles) and in the RK group (open circles). The correlation coefficient for the control group was 0.602 ($n = 30$, $P < 0.01$); the line drawn through the points was determined from the equation $y = 2.4x - 1.6$. In the RK group the correlation coefficient was 0.434 ($n = 24$, $P < 0.05$); the line drawn through the points was determined by the formula $y = 1.5x + 39$. The slopes of these two lines are significantly different ($P < 0.01$).

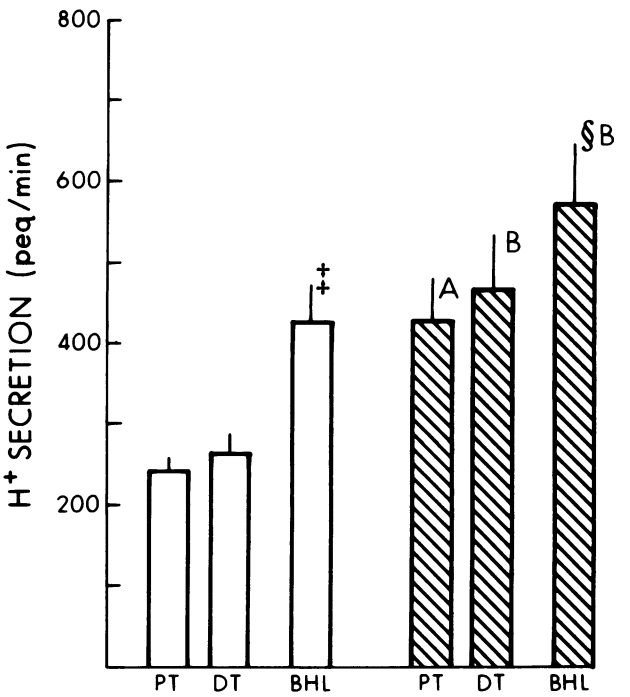

FIGURE 2 Hydrogen secretion before proximal (PT) and distal (DT) micropuncture sites compared with net secretion before the BHL of deep nephrons in the control group (open bars) and in the RK group (hatched bars). Values are mean ±SE. ‡, significant difference between the mean of BHL and PT or DT, $P < 0.005$, §, $P < 0.001$. A, significantly different from PT and DT in the control group at $P < 0.001$ and B, $P < 0.025$.

rising from 7.09±1.20 in controls to 31.1±5.0 peq/min in the RK group. The ammonium concentration in fluid obtained near the end of the proximal tubule of the RK averaged 4.17±0.38 mM, nearly twice the mean obtained in the CK. Absolute delivery of ammonium rose from 18.4±1.9 to 66.2±5.6 peq/min after renal mass was reduced by two-thirds, ($P < 0.001$). The increase was primarily a consequence of entry of ammonium between the glomerulus and the site of collection. This is evidenced by the proportional increase in $T_{NH_4^+}$ and the twofold rise in the fractional delivery of ammonium to the end of the proximal tubule. Further, this increased delivery was more than could be accounted for by the differences in tubule flow rate (Fig. 1). Despite the marked increase in delivery of buffered acid to the end of the proximal tubule of the RK, fractional bicarbonate reabsorption before this micropuncture site was reduced and absolute delivery of bicarbonate increased from 54.4±6.9 to 134±20 pmol/min. In fractional terms, delivery rose from 19.3±2.2 to 29.4±3.7% ($P < 0.005$). Despite the marked increase in buffered acid delivery to the end of the proximal tubule of the RK, total hydrogen secretion before this site did not increase in proportion

to the reduction in renal mass. The increase was less than twofold and averaged 433±54 peq/min in contrast to 238±16 peq/min for the CK (Fig. 2).

The in situ pH determined at end distal micropuncture sites was not significantly different in the two groups of animals. However, titratable acid concentration and delivery were strikingly greater in the RK group. Tubule fluid titratable acid concentration averaged 4.84±0.78 in contrast to 2.22±0.39 meq/liter in the control group. Titratable acid delivery was increased nearly sixfold, averaging 31.8±7.6 vs. 5.42±1.25 peq/min in sham-operated rats. Ammonium delivery to this site was also greater after renal mass was reduced, averaging 29.3±5.5 pmol/min in contrast to 10.7±1.9 pmol/min in the control group. However, as we have reported previously, in normal and acidotic rats (18, 19) delivery of ammonium to this site was significantly less than to the end of the proximal tubule. This is depicted graphically in Fig. 3. The net loss of ammonium between the end of the proximal and distal tubule averaged 38.2±5.0 pmol/min, fourfold greater than that of the sham-operated group, which was 9.1±2.3 pmol/min. As a consequence, fractional delivery of ammonium to the end distal site was less than to end proximal micropuncture sites in both groups of animals.

Bicarbonate delivery to the end of the distal tubule of the RK was markedly greater than to this site in shams. In absolute terms the mean value for the RK group was 58.8±12.5 pmol/min in contrast to 8.76±2.33 pmol/min in controls. Fractional delivery of bicarbonate rose from 4.16±1.29% in controls to 13.7±3.3% after renal mass was reduced. Hydrogen secretion prior to this site averaged 465±71 peq/min and did not differ significantly from the mean value obtained near the end of the proximal tubule (Fig. 2).

Deep nephrons. In the remnant kidney SNGFR measurements made near the bend of Henle's loop (BHL) of deep nephrons averaged 35.9±3.2 nl/min. This value is similar to what has been previously reported (22, 26) and was significantly greater than the mean obtained for the CK (Table IV). There was the expected increase in absolute and fractional delivery of fluid to the BHL after renal mass was reduced (21, 26). The tubule flow rate was greater for the RK and averaged 7.20±0.76, in contrast to 3.38±0.33 nl/min after sham operation ($P < 0.001$). Fractional fluid delivery increased from 14.5±0.9 in controls to 20.6±1.3% in this setting ($P < 0.001$).

Acid measurements were made in fluid obtained near the BHL of 35 deep nephrons in the sham-operated group and from 27 nephrons in the RK group. Mean values for these data are presented in Table IV and are contrasted with measurements obtained near the end of the proximal tubule of surface nephrons in Fig. 4. Mean tubule fluid pH measured near the BHL in the remnant group averaged 7.14±0.08. This value was significantly less than the mean for the controls

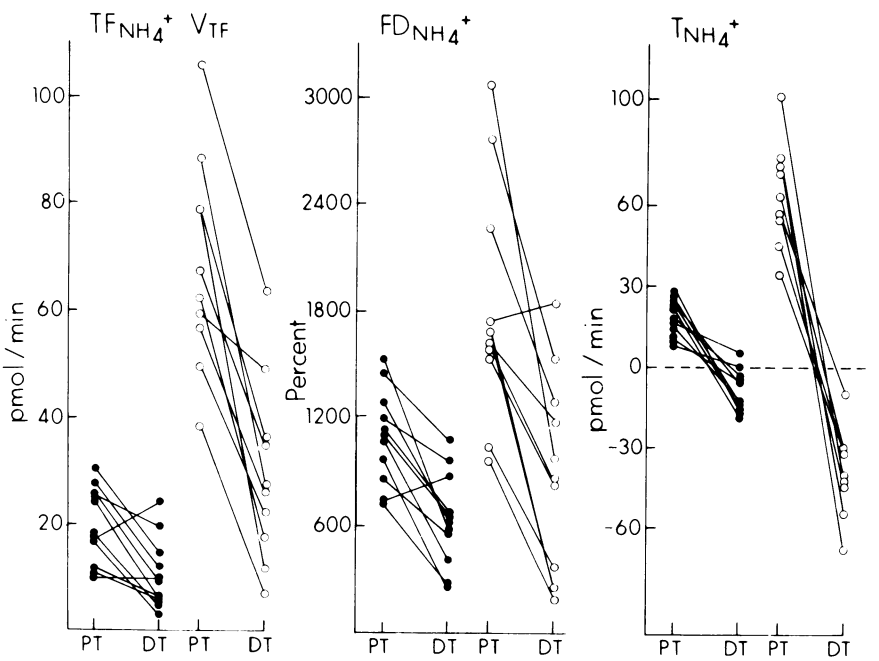

FIGURE 3 Comparison of absolute delivery of ($T_{NH_4^+}V_n$), fractional delivery ($FD_{NH_4^+}$), and entry of ammonia between the glomerulus and the end of the proximal and between this latter micropuncture site and the end of the distal tubule ($T_{NH_4^+}$). Closed circles, control group; open circles, RK group. Points represent the mean value for each rat.

TABLE IV

Effect of a Reduction in Renal Mass on Acid Delivery to the BHL of Deep Nephrons

Group	SNGFR	V_{tf}	TF_{OSM}	TF_{pH}	TF_{TA}	$TF_{TA}V_{tf}$	$FL_{NH_4^+}$	$TF_{NH_4^+}$	$TF_{NH_4^+}V_{tf}$	$T_{NH_4^+}$	$FD_{NH_4^+}$	$FL_{HCO_3^-}$	$TF_{HCO_3^-}$	$TF_{HCO_3^-}V_{tf}$	$FD_{HCO_3^-}$
	nl/min		*mosmol/kg H_2O*		*meq/liter*	*peq/min*	*pmol/min*	*mM*	*pmol/min*		*%*	*pmol/min*	*mM*	*pmol/min*	*%*
Control	23.8	3.38	1,288	7.34	0.44	1.63	2.82	14.5	45.6	40.8	1,567	469	24.6	79.5	17.6
n = 15	±2.2	±0.33	±95	±0.05	±0.12	±0.53	±0.27	±1.7	±7.0	±6.7	±180	±41	±1.8	±9.7	±1.4
Remnant	35.9	7.20	972	7.14	0.79	6.87	5.02	9.52	70.2	65.1	1,400	666	16.6	113	19.0
n = 11	±3.2	±0.76	±47	±0.08	±0.16	±2.55	±0.48	±1.08	±13.3	±12.9	±181	±64	±1.6	±11	±2.2
P	<0.005	<0.001	<0.01	<0.05	NS	<0.05	<0.001	<0.05	<0.05	<0.05	NS	<0.01	<0.005	<0.05	NS

Values are mean±SE. See Table III for abbreviations.

(7.34±0.05, $P < 0.05$). In both groups in situ pH measurements made at this site were consistently and significantly greater than the values obtained near the end of the proximal tubule of surface nephrons. The concentration of titratable acid measured near the BHL was considerably lower than values obtained at end proximal sites of surface nephrons, but was not different in the two groups of animals. Because of the difference in the rate of tubule flow, delivery of titratable acid was fourfold greater after renal mass was reduced than after sham operation (6.87±2.55 peq/min vs. 1.63±0.53 peq/min in the control group). In both groups titratable acid delivery was considerably greater to end proximal micropuncture sites of surface nephrons (Fig. 4).

As expected, the filtered load of ammonium in deep nephrons was significantly greater in the remnant group than in shams. The tubule fluid concentration of ammonium measured near the BHL was actually lower in the remnant group, averaging 9.52±1.08 mM compared with 14.5±1.7 mM after sham operation. Despite this, delivery of ammonium to the bend increased significantly, from 45.6±7.0 to 70.2±13.3 pmol/min. This increase was primarily due to the differences in nephron size resulting from hypertrophy of the RK. Thus, $T_{NH_4^+}$ was greater to this site in the RK but fractional delivery of ammonium to this site was unchanged.

Absolute bicarbonate delivery to the BHL was significantly greater after remnant formation, averaging 113±11 pmol/min in contrast to 79.5±9.7 pmol/min in the control group ($P < 0.05$). This occurred despite the fact that tubule fluid bicarbonate concentration was actually lower after renal mass was reduced, (16.6±1.6 vs. 24.6±1.8 mM in controls, $P < 0.005$). On the other hand, fractional delivery of bicarbonate to

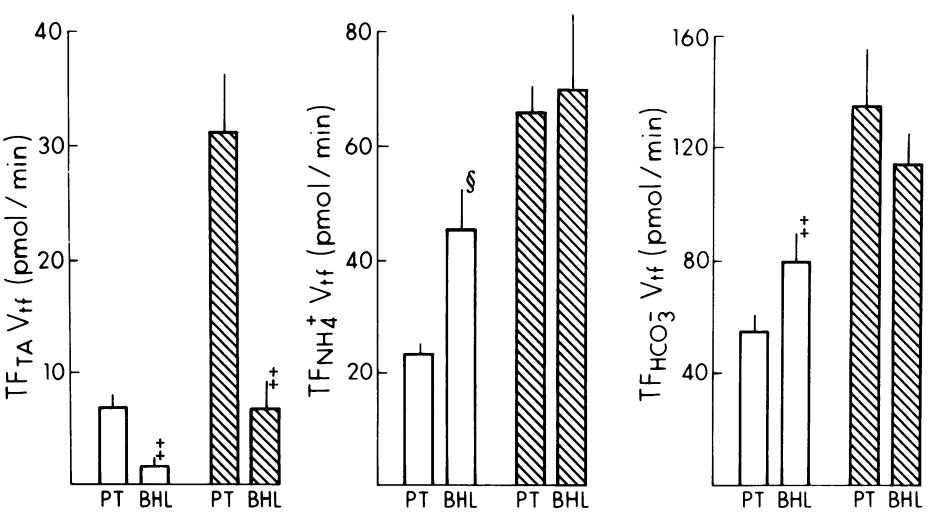

FIGURE 4 Delivery of titratable acid, ammonium, and bicarbonate to the end of the proximal tubule (PT) and the BHL after reduction in renal mass (hatched bars) and under control conditions (clear bars). Level of significance: ‡, $P < 0.005$; §, $P < 0.001$.

TABLE V
CD Function after Renal Mass is Reduced

	FD_{H_2O}		TF_{osmol}		TF_{pH}		TF_{TA}		TA index		$TF_{NH_4^+}$		$FD_{NH_4^+}$		NH_4 index		NA index	
	Prox	Tip	Prox	Tip	Prox	Tip	Prox	Tip	Prox	Tip	Prox	Tip	Prox	Tip	Prox	Tip	Prox	Tip
	%		*mosmol/kg H_2O*				*meq/liter*				*mM*		%					
Control group $n = 14$	1.27 ±0.25	0.78* ±0.07	1193 ±62	1360* ±77	6.24 ±0.10	5.62* ±0.10	50.5 ±5.2	67.0‡ ±6.9	0.57 ±0.11	0.43 ±0.03	105 ±11	142* ±11	902 ±96	862 ±94	1.05 ±0.09	1.00 ±0.09	1.60 ±0.14	1.46 ±0.09
Remnant group $n = 11$	3.42 ±0.25	2.81§ ±0.21	903 ±39	982§ ±34	6.08 ±0.11	5.63‡ ±0.11	47.8 ±5.0	56.2‖ ±6.0	1.57 ±0.17	1.52 ±0.16	60.1 ±6.8	74.6‖ ±9.1	1323 ±133	1335 ±144	1.93 ±0.17	2.01 ±0.24	3.48 ±0.29	3.58 ±0.36
P	<0.001	<0.001	<0.001	<0.001	NS	NS	NS	NS	<0.001	<0.001	<0.01	<0.001	<0.025	<0.01	<0.001	<0.001	<0.001	<0.001

Values are the mean±SE. See Table III and text for abbreviations. Significantly different from CD_{prox}:

* $P < 0.001$.

‡ $P < 0.01$.

§ $P < 0.005$.

‖ $P < 0.05$.

the BHL of deep nephrons did not differ significantly in the two groups of rats. Although total hydrogen secreted proximal to this site of collection was significantly greater in the RK group, averaging 624±70 peq/min vs. 432±42 peq/min (Fig. 2), the increase was not in proportion to the reduction in renal mass.

Papillary CD function

Mean measurements obtained near the base and tip of the CD of control and RK are presented in Table V. The distance between proximal and tip CD sites averaged 0.81±0.05 mm in the control group. This value was not statistically different from that of RK (0.79±0.03 mm). In both groups fractional delivery of fluid fell between the base and tip of the CD and was accompanied by a significant increase in the osmolality of fluid between these two sites. However, the fraction of the filtered load of fluid remaining at proximal and tip CD sites was greater for the RK than the CK. Similarly the osmolality of fluid obtained at the base of the CD was significantly less after renal infarction than after sham operation (903±39 vs. 1,193±62 mosmol/kg water in controls, $P < 0.001$). These results are similar to what we have previously reported after remnant formation and in hydropenia (22).

In situ pH measurements at proximal and tip CD sites were not statistically different in the two groups of animals. In both groups the pH measured at proximal sites was greater than the mean obtained at tip CD sites. The concentration of titratable acid at both sites was similar in the two groups of animals. The TA index averaged 0.57±0.11 at the proximal and 0.43±0.03 at tip CD sites after sham operation, which are similar to values that we have previously reported (19). The TA index measured at these two CD sites was threefold greater in the RK group. However, as in the control group there was no statistical difference in measurements made at these two sites of collection.

The ammonium concentration measured near the base of the CD was significantly less in the RK, averaging 60.1±6.8 mM in contrast to 105±11 mM after sham operation. In both groups, the concentration rose between the base and the tip of the CD. Fractional delivery of this buffer to the base of the CD was 902±96% after sham operation and did not change by tip CD sites. In the remnant group delivery was greater, averaging 1,323±133%, but as with shams, was not different from the average obtained near the tip of the CD. The relationship between fractional delivery of ammonium to the end of the proximal tubule and the base of the CD is depicted in Fig. 5 for both groups of animals. In controls, delivery to these sites did not differ statistically. However, $FD_{NH_4^+}$ to CD_{prox} in the RK group was significantly less than the mean measured near the end of the proximal tubule. The difference in delivery averaged 568±109% of the filtered ammonium in contrast to 93±53% found in the CK ($P < 0.005$). These differences are depicted graphically in Fig. 6. As expected the index for ammonium measured near the base of the CD was not different from the mean obtained at tip CD sites. Similarly in the RK group, the NH_4^+ index did not change statistically between base and tip CD sites. However, the values were significantly greater at both CD sites in the RK than in the CK group. The NA index measured

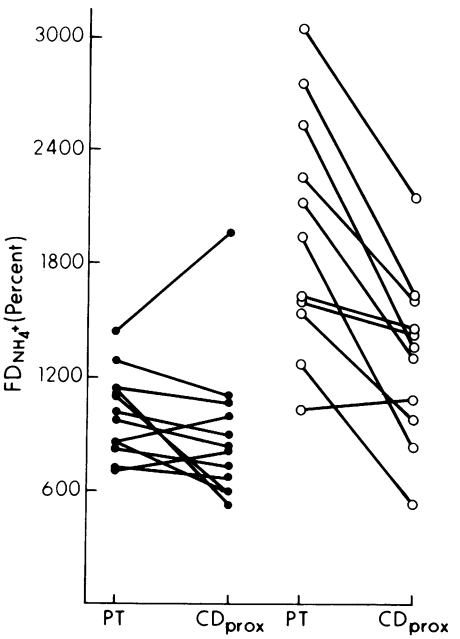

FIGURE 5 Comparison of fractional ammonium delivery to the end of the proximal tubule (PT) and CD_{prox} under control conditions (closed circles) and after a reduction in renal mass (open circles).

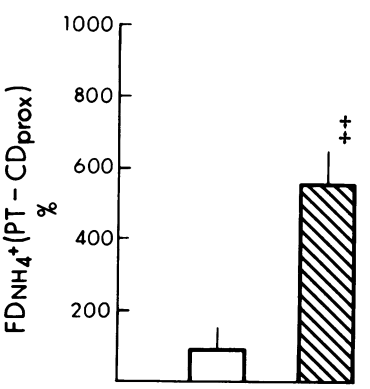

FIGURE 6 Difference in fractional delivery of ammonium to the end of the proximal tubule and the base of the CD in sham-operated rats (open bars) and after remnant formation (hatched bars). ‡, difference significant at $P < 0.005$.

near the base of the CD was also twofold greater after remnant formation. However as with controls, there was no change in this value along the terminal segment of the CD.

In Fig. 7 the mean Indices of buffered acid excretion measured near the end of the distal tubule of surface nephrons are compared with the average values obtained near the base of the CD in the two groups of animals. As expected, in both groups the NA index increased significantly between these two micropuncture sites; however, the increase was significantly greater after remnant formation, averaging 0.818±0.205 vs. 0.310±0.110 in controls ($P < 0.001$). In both groups the NH_4^+ index rose significantly between the end of the distal tubule and the base of the CD; however, the difference between the increases did not achieve statistical significance. Similarly, the increase in the TA index between the end of the distal tubule and the base of the CD in the remnant group was not statistically different from that of shams.

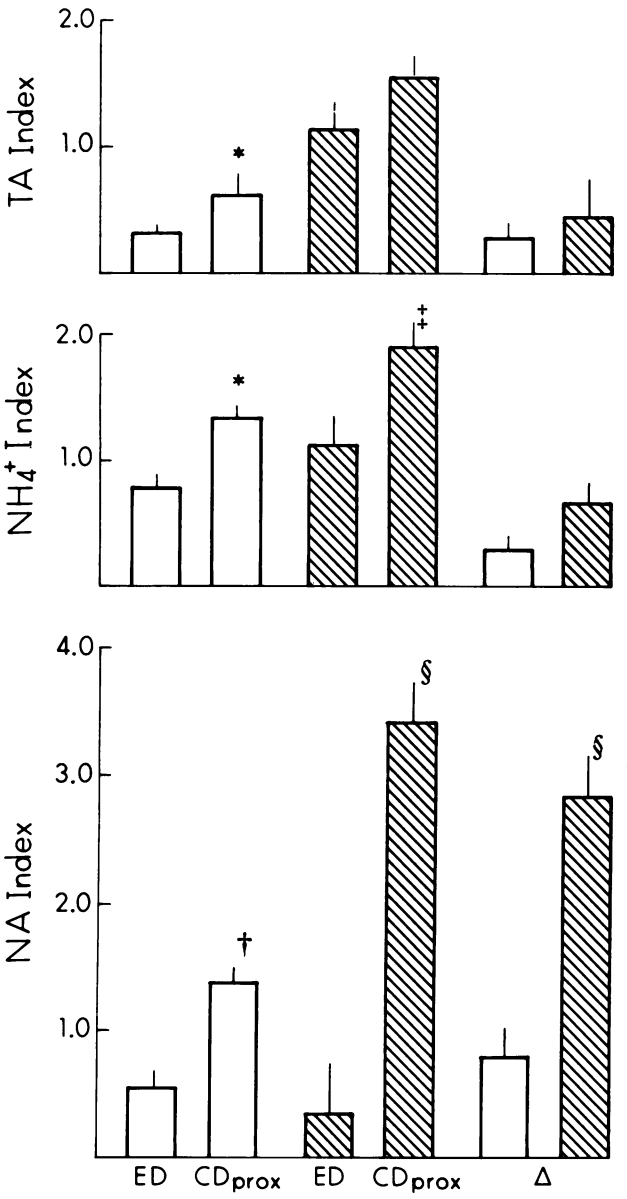

FIGURE 7 Indices of acid entry (mean±SE) obtained near the end of the distal tubule (ED) and near the base of the CD (CD_{prox}) after sham operation (open bars) and after remnant formation (hatched bars) Values are mean±SE. Δ is the mean±SE for the difference between CD_{prox} and ED. Values obtained at these sites and mean Δ values obtained in the group with the RK are significantly different at *, $P < 0.05$, †, $P < 0.01$, ‡, $P < 0.005$, and §, $P < 0.001$.

DISCUSSION

In this study a two-thirds reduction in renal mass affected the capacity of the residual functioning nephrons to excrete acid in ways similar to those described by other investigators in humans (4, 10–13) and in experimental animals (14–17). That is, the capacity of the RK to excrete acid was significantly impaired and a metabolic acidosis ensued. In this setting titratable acid excretion and urine pH measurements were statistically the same for the sham-operated group and the group with the RK. In contrast, ammonium excretion was reduced by nearly 50% after RK formation. However, as in previous studies (1–4, 10–17), there was clear evidence of enhanced ammonium production by the RK. That is, when factored by filtered load the excretion of ammonium from the RK was greater than from the CK (Table II). Therefore, it seems reasonable to assume that a partial infarction of the left kidney followed by right nephrectomy provides a valid model for the segmental analysis of the adaptive changes in acid excretion by superficial nephrons, deep nephrons, and the terminal segment of CD when renal mass is reduced.

After renal mass is reduced, ammonium delivery to the end of the superficial proximal tubule in the RK group rose more than threefold, averaging 66 pmol/min. Although this increase appeared to be proportional to the decrease in renal mass, it is difficult to assess the absolute contribution to ammonium excretion by superficial nephrons because the number remaining in the RK is uncertain. However, if one assumes that (*a*) nephron hypertrophy results in a 29% increase in the weight of the RK (27), (*b*) 72% of the nephrons remaining are superficial (28), and (*c*) the kidney of a rat contains 30,000 nephrons (29), then one can estimate the total number of surface nephrons contained in the remnant kidney from the following formula: $0.71(RK/CK) \times 21{,}600$, where RK is the mean weight of RK (268 mg) and CK is the mean weight of the left kidney (426 mg) of the control group of those rats that underwent micropuncture study.

Thus, ~9,700 surface nephrons are contained in the RK of the animals that underwent micropuncture study. If these estimates are correct, then 676±44 nmol/min are delivered out of the superficial proximal tubules of the RK. This value was not significantly different from that calculated for both kidneys of the sham-operated group (747±73 nmol/min). Although there was an increase in the filtered load of ammonium and in nephron size after RK formation, the major reason for the increased delivery was that entry of ammonia along the proximal tubule was markedly enhanced (Table III). Several factors appear responsible for this. First, the tubule fluid flow rate near the end of the proximal tubule of the RK was more than twice the control value (Table III). This increase would optimize the concentration gradient for the diffusion of ammonia into this tubule segment and therefore increase delivery of ammonium to the proximal micropuncture sites. Indeed, the flow dependence of ammonia entry along the proximal tubule is readily apparent in Fig. 1. Nevertheless, it does not seem likely that this increase in flow rate is the entire explanation, since the concentration of ammonium in fluid obtained at this site was higher in the RK than in the control group. Further, ammonia entry along the proximal tubule was greater in the RK group at any level of flow. A second factor is that luminal pH measured near the end of proximal tubule of the RK was slightly lower than that measured in controls. This increased pH gradient would favor luminal entry and entrapment of ammonia in the proximal tubule. A third factor that could have contributed to an enhanced entry of ammonia along the proximal tubule was the mild metabolic acidosis exhibited by the RK group. The mean arterial pH and plasma bicarbonate concentration were significantly lower in this group than in the sham-operated group. However, ammonium delivery out of the proximal tubule of the RK was substantially greater than that we have reported for rats chronically fed ammonium chloride (18, 19). In those studies, we found that ammonium delivery to the end of the proximal tubule increased by only twofold in face of metabolic acidosis that was more profound than that seen after RK formation. Indeed, the difference in delivery to this site under the two experimental conditions is more strikingly seen when ammonium delivery is factored by kidney weight. In this study, delivery to this micropuncture site in the RK group averaged 259±27, fivefold the value of 51.5±5.2 pmol/min per g kidney wt we reported for the group made chronically acidotic (19).[2] Thus, the changes in ammonium delivery seen after renal mass was reduced can be only partially explained on the basis of changes in the acid-base status of the rat. Nevertheless, it seems likely that an increase in the cortical production of ammonia is a major factor contributing to the increase in the end proximal delivery of ammonium. Although the rate of ammonium production cannot be determined from the data presented in this work, an estimate of differences in the production of ammonia by renal cortical tissue in the two groups of animals can be obtained by calculating the pNH_3 of the cortex. If it is true that ammonia entry into the tubule lumen is due to nonionic diffusion of the free base NH_3 where protonation and entrapment occurs (30), and that NH_3 is in equilibration throughout all structures located in the cortex (31, 32), then given the in situ pH and ammonium concentration of fluid obtained near the end of the proximal tubule, the pNH_3 of the renal cortex can be calculated with the following formula (33, 34):

$$pNH_3 = \frac{\text{total } NH_4^+}{10^{pK-pH} + 1} \times \frac{22.09}{\alpha}.$$

Using a pK of 9.02 (35) and the solubility coefficient (α) of 0.626 (34), cortical pNH_3 averaged 1,267±234 mmHg $\times 10^{-6}$ in the RK group, twofold the value in the sham-operated group (658±70 mmHg $\times 10^{-6}$). If intracellular pH fell during acidosis (36) and if ammonia entry was due solely to diffusion and no increase in synthesis occurred, then one would expect that the cortical pNH_3 of the RK group would be lower than that of the sham-operated group. The results of the present study are therefore most consistent with enhanced cortical production of ammonia by the RK, a finding that is in agreement with previous studies of this model of renal disease (17).

The rate of titratable acid excretion also increased in proportion to the decrease in renal mass; this is compatible with previous studies in which titratable acid excretion from the diseased kidney did not change (4, 11, 12). This increased excretion was associated with a similar increase in proximal delivery of titratable acid and was most likely a consequence of the different filtered loads of phosphate in the two groups of animals (4, 11, 12).

In the RK total hydrogen secretion before the end of the proximal tubule increased by less than twofold the value obtained in the sham-operated group. This increase was in near proportion to the adaptive change in SNGFR and was therefore not in proportion to the threefold decrease in renal mass (Fig. 2). Thus, bicarbonate delivery to the end of the proximal tubule was increased in both absolute and fractional terms. The finding that bicarbonate reabsorption in the proximal segments of surface nephrons is lower after renal mass is reduced is compatible with previous studies in humans (4, 5) and in experimental animals (6–8). The mechanism for this decreased reabsorption cannot be

[2] The mean value was derived from data presented in Tables II and IV of reference 19.

determined from the present study, although part of the explanation may relate to the decrease in fractional sodium reabsorption that occurs in this segment of the RK (1, 6, 22). As a result there was an increase in bicarbonate delivery to the distal tubule where the capacity to secrete hydrogen ion is more limited (37).

After renal mass was reduced delivery of ammonium to the BHL of deep nephrons increased but not to the same extent as ammonium delivery to the end of the proximal tubule of surface nephrons (Fig. 4). If the previously stated assumptions are correct, and ~3,770 deep nephrons are contained in the RK, then total delivery of ammonium out of the proximal segments of this nephron population would be 264±50 nmol/min. This is less than half of the ammonium delivered out of the superficial proximal tubules. In the sham-operated group total ammonium delivery to the BHL of deep nephrons and to the end of the proximal tubules of surface nephrons did not differ statistically (329±47 and 396±14 nmol/min, respectively). Thus, while there was evidence of an increase in ammonium production by the deep nephron proximal segments of the RK, total ammonium delivery to the BHL of all deep nephrons of this kidney did not approach the twofold increase needed for complete adaptation. These results are in sharp contrast to those observed in chronic metabolic acidosis (18, 19). In that setting, the deep nephron population played an important role in the renal response to an acid load. The ammonium concentration of loop fluid more than doubled and delivery to the BHL increased significantly. The mean ammonium delivery was 152±17 pmol/min per g kidney wt, more than threefold that obtained near the end of the proximal tubule of acidotic rats.[3] The response of deep nephrons to an acid load was therefore greater than that of surface nephrons (18, 19). In this study, when factored by kidney weight, delivery of ammonium to the deep nephron BHL of the RK was 260±38 pmol/mg per g kidney wt ($P < 0.025$) and was not different from the mean obtained near the end of the proximal tubule. This value was less than twice that obtained in chronic acidosis, whereas the delivery of ammonium to end proximal micropuncture sites after RK formation were more than five times that observed after the induction of a chronic metabolic acidosis.

The failure in complete adaptation by deep nephrons after a reduction in renal mass does not appear to be a consequence of altered proximal proton secretion; in this study hydrogen ion secretion in this segment of deep and surface nephrons increased to a similar extent (Fig. 2). Rather, it seems that the differences in delivery under the two experimental circumstances are related to the reduction in the solute concentration of the papillary interstitium. Several factors support this hypothesis. First, the osmolality of loop fluid was significantly less after RK formation than after sham operation (Table IV) or after the induction of a chronic metabolic acidosis (18). Second, the concentration gradient for ammonium between the end of the proximal tubule and the BHL (4.8 to 9.4 mM) was considerably less in the RK than in the CK (2.5 vs. 14.8 mM near the BHL). Indeed, the concentration of ammonium in fluid obtained near the BHL was actually lower in the RK group than in controls. Finally, estimates of the pNH_3 content of the papillary interstitium averaged 4,963±764 mmHg $\times 10^{-6}$ in the RK group, a value which was less than half the mean of 11,754±2,425 mmHg $\times 10^{-6}$ calculated for the control group. The countercurrent multiplication of ammonia in the papillary interstitium appears impaired when renal mass is reduced. One factor responsible for this failure is that the tubule flow rate measured near the BHL of deep nephrons is increased. An increase in medullary blood flow to the papilla would also serve to dissipate the gradient, although to date there is no information available to support this possibility. Such changes in the dynamics of countercurrent multiplication might enhance the upstream movement of ammonia out of the renal tubule of deep nephrons and account for the smaller increase in ammonium delivery to BHL than to end proximal micropuncture sites.

In situ pH measurements made along the distal tubule of the RK averaging 6.61, were not significantly different from values measured at this site in sham-operated rats, and the gradient for hydrogen secretion in the distal tubule of the RK is the same as in controls. Because of the difference in flow rate, however, free hydrogen delivery to the end of the distal tubule of the RK is actually greater. Nevertheless, these data suggest that the segments between the end of the proximal and distal tubule of surface nephrons are not major sites of proton secretion under normal conditions or after renal mass is reduced. That is, the rate of hydrogen ion secretion that occurred between the end of the proximal and distal tubule in both groups of animals was not great enough to be detected with the methods used in the present work (Fig. 2). Nevertheless, distal delivery of bicarbonate was less than to end proximal micropuncture sites. It seems likely that part of this decrease was due to bicarbonate buffering by the hydrogen ion previously bound to the ammonia, which was lost in the intervening segment, and to continued hydrogen secretion along the remaining proximal segments of the nephron.

In this study there is evidence to suggest than when renal mass is reduced ammonium entry along the medullary and terminal segments of the CD is inappro-

[3] The mean value was derived from data presented in Tables II and V of reference 19.

priately low. Although the index for net acid excretion increased to a greater extent between the end of the distal tubule and CD_{prox} in the group with the RK, it is apparent that the greater increase in this index was not a reflection of differences in ammonium entry or in the titration of phosphate along this segment (Fig. 7). Rather, the increase seems attributable to the titration of the bicarbonate delivered out of the distal tubule. Thus, a reduction in renal mass does not result in enhanced entrapment of ammonia along this segment. These data contrast sharply with those obtained in rats made chronically acidotic (18, 19). In those studies, the increase in the NH_4^+ index between the end of the distal tubule and the base of the CD was twofold greater after the chronic administration of ammonium chloride. In fractional terms, >800% of the filtered ammonium entered along this segment in contrast to <300% in the control group. Further, fractional delivery of ammonium to the base of the CD was significantly greater than to the end of the proximal tubule in chronically acidotic rats (18), whereas in the present study delivery of ammonium to the base of CD was consistently lower in the group with the RK (Figs. 5 and 6).

Although the entire explanation for this failure to reentrap ammonia along the CD after RK formation remains unclear, several factors are apparent. First, as in controls, the terminal segment of the CD of the RK secretes hydrogen ion. Tubule fluid pH measured at the base of the papilla was significantly greater than that measured near the tip. The decline in luminal pH between the end of the distal tubule and the base of the CD was also similar in the two groups of animals, which suggests that the gradient for hydrogen ion secretion along the entire CD is the same in both groups of animals. Therefore, an alteration in the hydrogen ion gradient between the lumen and cell of the CD cannot be an explanation for the loss of ammonium produced in the proximal tubule. Second, as in acidosis (18, 19), the loss of ammonia between proximal and distal micropuncture sites is enhanced after renal mass is reduced (Fig. 3 and Table III). Thus, delivery of ammonia to the medullary interstitium appears appropriate after a reduction in renal mass. On the other hand, it seems likely that a significant portion of the ammonia produced in the proximal segments of surface nephrons is lost to the circulation. The concentration of ammonium in the tubule fluid obtained near the BHL was lower after RK formation than after sham operation, and delivery of ammonium to this site did not increase appropriately. Furthermore, Maclean and Hayslett (17), comparing control and RK groups found that there was a significantly greater percentage of the ammonium produced by the RK in the venous effluent than in the final urine. Thus, it seems likely that the altered dynamics of solute concentration in the papillary interstitium after renal mass is reduced may affect the capacity of the RK to establish high concentrations of ammonia in the papilla. This would decrease the gradient favoring the movement of ammonia into CD fluid.

In conclusion, the present study suggests that two mechanisms underly the incapacity of the RK to secrete a hydrogen load. First, hydrogen secretion in the proximal segment of deep and surface nephrons does not increase in proportion to the decrease in renal mass. As a consequence, more bicarbonate is delivered out of these segments. This obligates the distal nephron segment to reclaim bicarbonate rather than to titrate nonabsorbable buffers. Second, the decrease in ammonium excretion after renal mass is reduced is due to a failure to reentrap ammonia along the distal nephron rather than to a reduction in the capacity of the cortical segments of the nephron to produce ammonia.

ACKNOWLEDGMENTS

We thank Dr. Saulo Klahr, Director of the Renal Division of Washington University School of Medicine, St. Louis, MO, for critically reading this manuscript and Dr. N. Carter and Dr. L. Pucacco of the University of Texas Health Science Center, Dallas, TX, for their kind assistance in developing the technique for manufacturing pH microelectrodes in our laboratory. We also express our appreciation to John Fritsch for his excellent technical assistance, and to James Havranek, Helen Odle, and Karon Farris for their help in the final preparation of this manuscript.

These studies were supported by grants from the American Heart Association (77-885) and the U. S. Public Health Service (AM-00976 and AM-07126).

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