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ORIGINAL PAPER

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Angiotensin II involvement in adaptive enteric oxalate excretion in rats with chronic renal failure induced by hyperoxaluria

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Abstract Enteric secretion of oxalate is induced in rats that have chronic renal failure produced by 5/6 nephrectomy [2]. The purpose of the present study was to examine renal and intestinal handling of oxalate in rats with chronic renal failure (CRF) induced by chronic hyperoxaluria. A rat model for chronic renal failure, induced by chronic hyperoxaluria (CH-CRF), was produced by unilateral nephrectomy combined with dietary ethylene glycol for 4 weeks. Both intact and unilateral nephrectomized rats (UN) without the oxalate load served as controls. Renal handling of oxalate was assessed by measurement of renal clearance of oxalate and creatinine while colonic handling of oxalate and chloride was determined by in vitro transepithelial flux measurements. Angiotensin II mediation was assessed by sensitivity of the transport processes to the AT_1 receptor antagonist losartan. Renal and colonic handling of oxalate in UN rats were similar to intact controls. The CH-CRF rats were hyperoxalemic, hyperoxaluric, and exhibited a twofold increase in oxalate clearance despite a 50% drop in creatinine clearance. Distal (but not proximal) colonic handling of oxalate in CH-CRF rats was reversed from net oxalate absorption seen in UN and intact controls to net secretion that was sensitive to losartan in vitro. Conclusion: Although enteric oxalate secretion can be correlated with elevations in plasma oxalate in the absence of overt renal insufficiency by an ANG II-independent mechanism, the present results suggest that some degree of renal insufficiency is necessary to induce ANG II-mediated colonic oxalate secretion.

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Introduction

Enteric elimination of oxalate has been shown to occur in rats with chronic renal failure (CRF) induced by surgically removing 5/6 of the renal mass [1, 2]. It was evident from one study, which followed the fate of ^{14}C-oxalate infused subcutaneously by a mini-osmotic pump, that fecal excretion of the tracer was fivefold higher in CRF rats when compared to normal rats [1]. Subsequently, we directly examined the magnitude and direction of the unidirectional oxalate fluxes across isolated intestinal segments removed from a similar CRF rat model. We found that the large intestine is the primary site for an adaptation in oxalate handling in CRF where basal oxalate absorption is reversed to secretion by significant changes in both unidirectional fluxes [2]. Furthermore, this adaptive secretory response in renal insufficiency involves angiotensin II (ANG II) receptor agonism via an AT_1 receptor subtype, as it can be inhibited by AT_1 receptor antagonists [3]. It was also demonstrated that the adaptive response of colonic oxalate secretion in the CRF rat could be simulated in control rat distal colon in vitro by ANG II receptor agonism [3]. Together, these observations imply that ANG II plays a role in local modulation of adaptive enteric oxalate excretion.

Since these previous studies involved rats with chronic renal failure induced by 5/6 nephrectomy, the present study addressed the questions of whether enteric oxalate elimination occurs in an animal model with chronic renal failure induced by chronic hyperoxaluria (CH-CRF) and, if it does, whether this response is mediated via ANG II receptor action. The present studies show that both renal secretion and enteric secretion of oxalate are initiated in this CRF model and further suggest that the changes in active intestinal transport are confined to the distal segment of the colon

and are, at least partly, mediated by AT_1 receptor agonism.

Methods

Animals

Male Sprague Dawley rats (285–325 g) were used in the following studies. The rats had free access to drinking water and Purina rat chow 5001. All animal experimentation described in this article was conducted in accord with the University of Florida and the NIH Guide for the Care and Use of Laboratory Animals. In order to produce oxalate-induced chronic renal failure, a unilateral nephrectomy was performed on each animal followed by 4 weeks of ethylene glycol treatment (0.75%, administered in the drinking water). Briefly, general anesthesia was induced with an intraperitoneal injection of pentobarbital (40 mg/kg body weight) and the surgical procedure of a right nephrectomy was performed extraperitoneally under aseptic conditions. Prior to initiating the treatment with ethylene glycol, the rats were given 1 week to recover from the surgery. Nontreated, unilateral nephrectomized rats (UN) served as controls. At the end of the specified treatment, prior to collecting blood and colonic tissues for the flux studies, urine from all rats was collected into vessels containing 3.0 ml of 3.5 N HCl over a period of 24 h. The animals were euthanized by an intraperitoneal injection of sodium pentobarbital (150 mg/kg body weight) and promptly exsanguinated by cardiac puncture. The blood was handled immediately with the appropriate precautions to prevent oxalogenesis [4]. Oxalate was measured in plasma and urine using a coupled enzymatic (oxalate decarboxylase and formate dehydrogenase) assay procedure routine in our laboratory [4]. Creatinine was determined in the urine and plasma samples using the Sigma kit assay 555A (Sigma Chemical Co., St. Louis, MO).

Flux studies

Flux experiments were conducted using both proximal and distal colonic tissues removed from control rats with two intact kidneys, UN rats, and CH-CRF rats, with the latter two groups being euthanized 5 weeks after surgery. As described previously [2], flat sheets of intestinal tissue were mounted in modified Ussing chambers with an exposed tissue area of 0.64 cm^2 and bathed on both sides by 10 ml of standard saline solution at 37°C, which was vigorously circulated by bubbling with a gas mixture of 95% O_2/5% CO_2. The standard saline contained the following solutes (millimole/liter): 139.4, Na^+; 5.4, K^+;1.2 Ca^{2+}; 1.2, Mg^{2+}; 123.2, Cl^-; 21.0, HCO_3^-; 0.6, $H_2PO_4^-$; 2.4, HPO^{2-}; and 10, glucose. Unidirectional fluxes of oxalate and chloride were measured using ^{14}C-oxalate and ^{36}Cl. The magnitude and direction of the net flux (J_{Net}) was determined by calculating the difference between the two unidirectional fluxes (mucosal to serosal, J_{ms} and serosal to mucosal, J_{sm}) measured for a control period of 45 min (Per I) at 15-min intervals, under short-circuit conditions. The net tracer fluxes were determined on matched tissues ($G_T \pm 20\%$) and the flux results are given for the number (n) of matched tissue pairs. Per I was followed by a second 45-min flux period (Per II) in order to determine either time-dependent effects or the effects of drug (losartan) addition. In previous studies, we determined that losartan at 10^{-3} M produced the maximal effect on chloride transport across the distal colon [5] and this concentration has been employed here. The electrical parameters of the tissue were also recorded at 15-min intervals throughout the entire experiment. Tissue conductance (G_T, mS·cm^{-2}) was calculated as the ratio of the open-circuit potential (mV) to the short-circuit current (I_{sc}, μA·cm^{-2}). ^{14}C-oxalate and ^{36}Cl were obtained from New England Nuclear (Boston, MA). The AT_1 receptor antagonist, losartan, was a gift from Merck & Co., Inc. (Rahway, NJ), and all other reagents were purchased from Sigma Chemical Co. (St. Louis, MO).

Statistical analysis

Results are presented as the mean ± one standard error (SE) for n tissues or tissue pairs. Statistical comparisons of more than two means were performed by a one-way analysis of variance (ANOVA) followed by Bonferroni's t-test for multiple comparisons with the control group. A paired t-test was used for the comparison of two means. In both cases,differences were considered significant if $p \leq 0.05$.

Results

Plasma and urine creatinine and oxalate concentrations

The results presented in Table 1 confirm a significant decrease in renal function in the CH-CRF group at the time the flux studies were conducted. Plasma creatinine was increased twofold and creatinine clearance was reduced by about 50% in the ethylene glycol-treated (CH-CRF) rats compared to both the nontreated (UN) group and a group of normal controls with both kidneys intact. It is also apparent from these results that the rats with one kidney have normal renal function.

Compared to humans, rats have a higher plasma oxalate concentrations and they excrete more oxalate in urine on a kilogram basis, as shown here and elsewhere [1, 2, 6]. The results in Table 2 show that, like creatinine, plasma oxalate and the renal handling of oxalate (as judged by urinary excretion, renal clearance, and oxalate/creatinine renal clearance ratios) are comparable in rats with one kidney (UN) when compared with controls (both kidneys intact). A mean oxalate/creatinine clearance ratio less than 1 indicates tubular reabsorption of oxalate occurring in both the UN and the control group. In contrast, renal handling of oxalate in the CH-CRF group was significantly altered due to the increased endogenous synthesis of oxalate and these rats were both severely hyperoxaluric and hyperoxalemic. Renal clearance of oxalate was increased twofold in the CH-CRF group in spite of a 50% reduction in renal clearance of creatinine and, as indicated by the mean oxalate/creatinine clearance ratio, the oxalate-loaded animal supported a significant renal secretion of oxalate.

Table 1 Plasma creatinine and urinary creatinine excretion in control, UN, and CH-CRF rats

Group	Plasma (mM)	Excretion (μmole/24 h)	Clearance (ml/min)	Urine Volume (ml/24 h)
Control ($n=14$)	0.05 ± 0.001	168.1 ± 12.5	2.41 ± 0.30	16.5 ± 2.25
UN ($n=8$)	0.05 ± 0.002	166.8 ± 7.7	2.34 ± 0.18	17.8 ± 1.7
CH-CRF ($n=7$)	0.10 ± 0.005*	162.6 ± 7.5	1.19 ± 0.09*	44.8 ± 1.9*

* An asterisk indicates a significant difference from the control group.

Table 2 Plasma oxalate and urinary oxalate excretion in control, UN, and CH-CRF rats

Group	Plasma (μM)	Excretion (μmole/24 h)	Clearance (ml/min)	Ox/Cr ratio
Control ($n=14$)	6.4 ± 1.1	10.5 ± 1.3	1.65 ± 0.41	0.67 ± 0.14
UN ($n=8$)	6.8 ± 1.2	10.1 ± 1.2	1.38 ± 0.33	0.62 ± 0.21
CH-CRF ($n=7$)	46.6 ± 8.4*	144.4 ± 6.1*	2.76 ± 0.67	2.51 ± 0.70*

*An asterisk indicates a significant difference from the control group.

Colonic oxalate and chloride transport

The results of the transport studies show that, similar to normal control rats with both kidneys intact, the distal colon of UN rats supports a significant basal net absorptive flux of oxalate and chloride (Fig. 1). The unidirectional and net fluxes, as well as the electrical characteristics (data not shown), of this segment were comparable in both UN and controls. In contrast, a reversal in the direction of the net oxalate flux was evident in the CH-CRF rats (unilateral nephrectomized treated with ethylene glycol) and this net secretion of oxalate occurred by way of significant alterations in both of the unidirectional fluxes. A significant reduction in the absorptive component of the chloride flux (J_{ms}^{Cl}) resulted in markedly diminishing net chloride absorption, but it is noteworthy that net chloride secretion was not stimulated in these tissues. These changes in fluxes were not accompanied by significant alterations in I_{sc} or G_t (data not shown).

Parallel studies of oxalate and chloride transport across the proximal colonic segment removed from the three rat groups confirmed a basal net secretion of both oxalate and chloride in both the control and UN groups with normal renal function (Fig. 2). Unlike the distal segment, however, there were no alterations in the unidirectional or net fluxes of oxalate and chloride in the

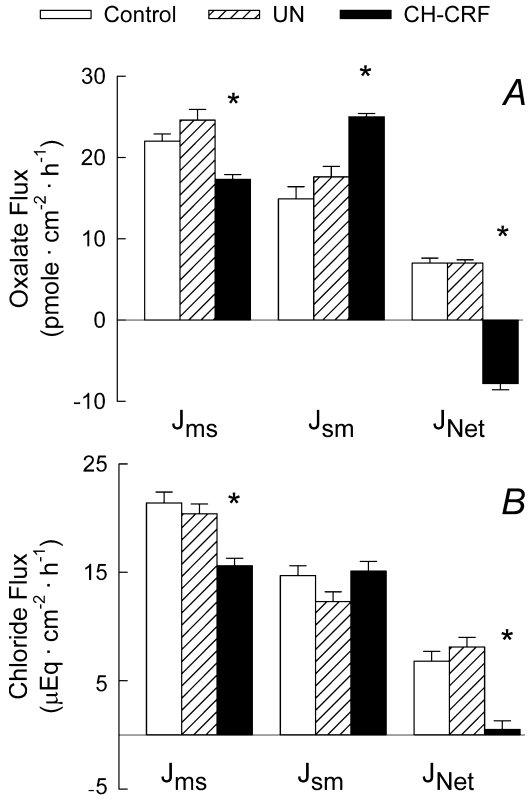

Fig. 1A, B Unidirectional and net transepithelial fluxes of oxalate (**A**) and chloride (**B**) across isolated, short-circuited segments of the distal colon ($n=7$ for each group) from control (*open rectangles*), unilateral-nephrectomized (*hatched rectangles*), and CH-CRF (*solid box*) rats. J_{ms} indicates mucosal to serosal ion flux; J_{sm} indicates serosal to mucosal flux; J_{net} is the calculated difference between J_{ms} and J_{sm}. An *asterisk* indicates a significant difference from the control group. Short-circuit current (I_{sc}) in controls was 4.1 ± 0.5 μEq·cm^{-2}·h^{-1} and was not significantly affected in UN or CH-CRF rats. Similarly, transepithelial conductance in the experimental groups was not significantly different from that of control tissues ($G_T = 9.9 \pm 0.5$ mS·cm^{-2})

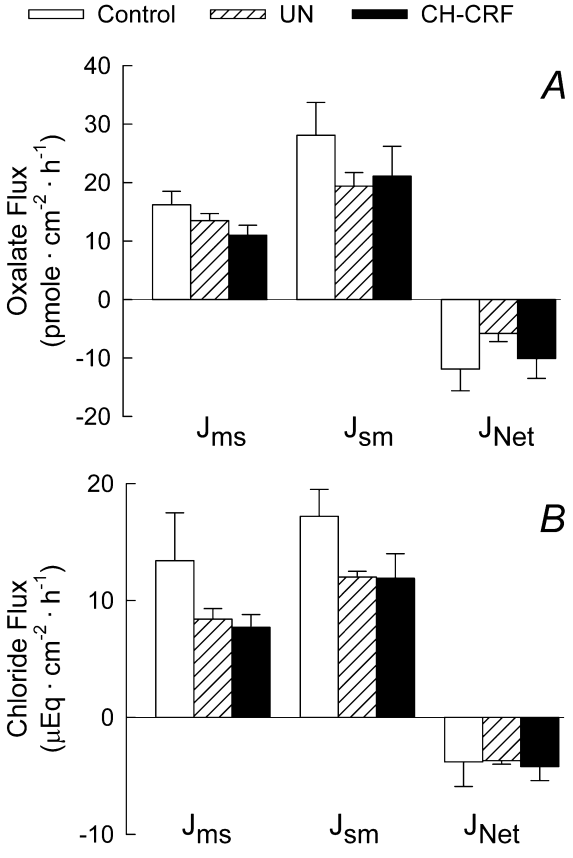

Fig. 2A, B Unidirectional and net transepithelial fluxes of oxalate (**A**) and chloride (**B**) across isolated, short-circuited segments ($n=5$ for each group) of the proximal colon from control (*open box*), unilateral-nephrectomized (*hatched rectangles*), and CH-CRF (*solid rectangles*) rats. J_{ms} indicates mucosal to serosal ion flux; J_{sm} indicates serosal to mucosal flux; J_{net} is the calculated difference between J_{ms} and J_{sm}. An *asterisk* indicates a significant difference from the control group. Control I_{sc} was 3.9 ± 0.4 μEq·cm^{-2}·h^{-1} and was not significantly affected in UN or CH-CRF rats. Similarly, transepithelial conductance in the experimental groups was not significantly different from that of control tissues ($G_T = 15.3 \pm 1.2$ mS·cm^{-2})

CH-CRF group. In addition, there were no changes in the associated electrical parameters (data not shown).

In all of these experimental series using distal and proximal colonic segments (Figs. 1 and 2), the results presented here were acquired during the first acquisition period (Per I). Results acquired during Per II showed that there were no significant time-dependent changes in either the fluxes or the associated electrical parameters in any series over the duration of two flux periods (data not shown). In addition, fluxes across proximal colonic tissues were not significantly altered by any of the in vivo or in vitro treatments employed in this study; consequently, the data are not presented for each experimental series that was conducted in parallel using proximal colonic tissue preparations.

Ethylene glycol, per se, could induce the alterations in oxalate transport observed in the ethylene glycol-treated rats. To test this possibility, ethylene glycol was added directly to the solution bathing the in vitro tissue preparations (proximal and distal colon) removed from healthy control rats to achieve a final concentration of 0.75% in the serosal and mucosal compartments. It was observed that exogenous ethylene glycol, added immediately after Per I, had no effect on the fluxes of either anion during Per II and the electrical characteristics of these tissues were also unchanged (data not shown).

The time course of the alterations in oxalate fluxes in the CH-CRF group was examined in the next experimental series and are depicted in Fig. 3. Distal colonic tissues were removed from unilaterally nephrectomized rats after 5 and 14 days of ethylene glycol treatment. For the purpose of comparison, the fluxes of UN (nontreated) and CH-CRF (30 days of treatment) are included (from Fig. 1). The results show that by day 5, the normal net absorptive flux of oxalate across this segment is already significantly reduced, and after 14 days net oxalate secretion has been initiated. The alteration in oxalate transport was accompanied by a reduction in net chloride flux, which was evident by day 14 (by coordinated changes in the unidirectional fluxes) and by day 30, net chloride absorption was negligible. On day 5, when a significant change in oxalate transport was observed, both mean urinary oxalate excretion (at 103 ± 11 μmole/24 h) and mean plasma oxalate concentrations (at 48 ± 14 μM) were already elevated ten times and seven times normal, respectively. In addition, mean plasma creatinine was concomitantly elevated to 0.08 ± 0.003 mM.

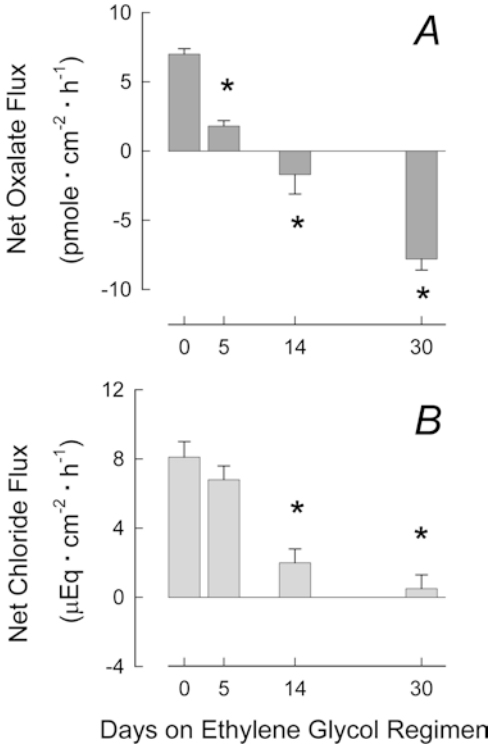

Fig. 3A, B Time course of adaptations in the net transepithelial fluxes of oxalate (**A**) and chloride (**B**) across isolated, short-circuited segments of the distal colon from control rats and unilateral-nephrectomized rats administered 0.75% ethylene glycol for 30 days. J_{ms} indicates mucosal to serosal ion flux; J_{sm} indicates serosal to mucosal flux; J_{net} is the calculated difference between J_{ms} and J_{sm}. For the purpose of comparison, the fluxes of UN (nontreated) and CH-CRF (30 days of treatment) are included (from Fig. 1). Sample sizes of 7, 9, 5, and 6 were employed at times 0, 5, 14, and 30 days, respectively. An *asterisk* indicates a significant difference from the control group (time zero)

Effects of AT_1 receptor antagonism on colonic anion fluxes

In a previous study involving rats with chronic renal failure induced by 5/6 nephrectomy (without ethylene glycol treatment), it was shown that losartan treatment of these CRF rats resulted in normalizing oxalate transport across the distal colon, indicating that ANG II was involved in mediating CRF-induced oxalate secretion [3]. Acute, in vitro antagonism of the CH-CRF secretory response (an increase in J_{sm}^{Ox} and J_{sm}^{Cl}) was examined in a separate experimental series. Losartan, at a concentration (10^{-3} M) known to have a maximum inhibitory effect on both K^+ and Cl^- secretion across this tissue preparation [5, 7], was added to the serosal solution bathing the CH-CRF tissue preparation in vitro, and a reduction in the secretory component of both anions was observed (Fig. 4). These results suggest that ANG II is involved, directly or indirectly, in mediating the oxalate secretory process in CH-CRF.

Discussion

Adaptive enteric oxalate excretion by the large intestine can have an important role in mass balance of oxalate, particularly in chronic renal failure. Three key pieces of information have emerged from our recent studies of colonic oxalate transport in rats with chronic renal failure induced by surgically removing 5/6 of the renal mass. First, the large intestine is the primary site for the

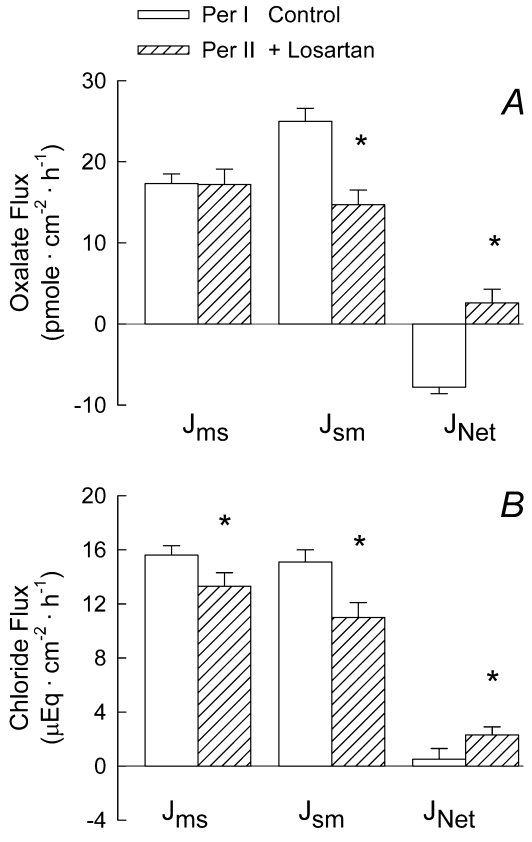

Fig. 4A, B Acute effects of AT_1 receptor antagonism on the unidirectional and net transepithelial fluxes of oxalate (**A**) and chloride (**B**) across isolated, short-circuited segments ($n=6$) of distal colon from CH-CRF rats. J_{ms} indicates mucosal to serosal ion flux; J_{sm} indicates serosal to mucosal flux; J_{net} is the calculated difference between J_{ms} and J_{sm}. Control fluxes were established in Period I (open rectangles), after which losartan was added to the serosal side at a final concentration of 1.0 mM. Period II measurements (hatched rectangles) commenced 15 min thereafter. An *asterisk* indicates a significant difference from the control group. Short-circuit current was significantly reduced (5.6 ± 0.7 to 4.6 ± 0.6 $\mu Eq \cdot cm^{-2} \cdot h^{-1}$) but G_T remained unchanged (12.1 ± 0.3 $mS \cdot cm^{-2}$)

CRF-induced adaptation where basal oxalate absorption is reversed to a net secretory flux [2] that is inhibited by AT_1 receptor antagonists [3]. Second, in CRF rat intestine, AT_1 receptors are up-regulated exclusively in the large intestinal segment [8]. Third, the effects of AT_1 receptor agonism and antagonism on oxalate transport in the CRF rat can be simulated in vitro in a control rat, which normally supports a basal absorptive flux of oxalate [3]. However, it has not been established, nor can it be assumed, that all rat models of chronic renal failure will behave identically in terms of renal and intestinal handling of solutes or in the regulation of epithelial solute transport, especially when challenged with an oxalate load. It is particularly important to note that the 5/6 nephrectomized rat model excreted normal amounts of urinary oxalate, and we observed only a modest increase in plasma oxalate (to ~12 μM), which was a direct result of reduced renal mass/function since these were not oxalate-loaded rats [2]. Consequently, this model does not reflect the severe hyperoxalemia coupled with hyperoxaluria that is apparent in conditions like primary hyperoxaluria as considered below.

The oxalate-induced CRF (CH-CRF) rat model

The present study employed a different CRF rat model from the one we have previously used [2] and it was established in an effort to simulate some of the clinical characteristics associated with PH and oxalate-induced renal failure. In the CH-CRF rat model, endogenous production of oxalate (from ethylene glycol metabolism) generated both marked hyperoxalemia and hyperoxaluria and induced chronic renal failure in a short period of time in rats with one intact kidney. It is notable that renal function and, in particular, renal handling of oxalate were not altered by unilateral nephrectomy alone, as we have shown here in rats for the first time. It should also be noted that renal function in ethylene glycol (0.75%) treated rats, with two intact kidneys, can remain unchanged for many weeks or months [6]. Consequently, in a pilot study prior to the present report, we determined that 4 weeks of treating unilateral nephrectomized rats with 0.75% ethylene glycol resulted in a twofold increase in plasma creatinine, which was comparable to that reported for the studies conducted using the 5/6 nephrectomized rat model [2]. The sevenfold increase in plasma oxalate concentrations to (~47 μM) in the CH-CRF rat model compared with basal levels found in normal rats is well within the range of plasma oxalate concentrations observed in some patients with PH, which can reach values up to, or in excess of, 100 μM [9, 10]. Likewise, the approximately14-fold increase in mean urinary oxalate excretion in CH-CRF reflects increments seen in oxalate excretion in PH. Compared with normal urinary values up to 0.4 mmole/24 h, urinary oxalate is commonly increased to 2–6 mmole/24 h in patients with PH [11]. Thus, the CH-CRF rat model generates degrees of hyperoxaluria and hyperoxalemia reminiscent of those found in PH patients.

Renal handling of oxalate in CH-CRF

Earlier investigations of renal mechanisms for oxalate transport by the rat kidney showed that oxalate is freely filtered at the glomerulus and undergoes bidirectional transport in the tubules. These studies also showed that oxalate clearance exceeds creatinine clearance, which is indicative of net tubular secretion of oxalate [12, 13, 14]. Our studies using a variety of rat models with and without renal insufficiency [2, 6] have yielded useful information regarding renal handling of oxalate. In 5/6 nephrectomized CRF rats we found that renal clearance of oxalate was reduced by 50%, which was accompanied by a modest increase in plasma oxalate concentration,

but these rats excreted normal amounts of urinary oxalate [2]. Studies in our laboratory examining both oxalate and creatinine clearances have revealed oxalate/creatinine clearance ratios less than 1 in the majority of control (89%) and 5/6 nephrectomized CRF rats (78%) [2]. We have also observed that renal secretion and/or reabsorption of oxalate (as judged by the clearance ratio) can occur in the presence or absence of renal failure [2], which suggests that renal transport of oxalate is under regulation, like the transport of other solutes and major plasma ions. Clearly, and not surprisingly, oxalate loading induces renal oxalate secretion, as a clearance ratio greater than 1 was observed in all of the CH-CRF rats and the mean clearance ratio (oxalate/creatinine) was increased fourfold over baseline values found in both control and UN groups. By comparison, however, in rats with normal renal function, the capacity of the kidney to secrete oxalate was found to be considerably greater and we have reported a mean clearance ratio 19 times normal in rats (with intact kidneys) administered oxalate in the food [6]. Thus, it appears that with the onset of renal failure the capacity for renal secretion of oxalate is severely diminished.

Enteric handling of oxalate vs chloride in CH-CRF

Our studies of both the rabbit and rat intestine have clearly shown that these epithelia exhibit the capacity to both absorb and secrete oxalate [15, 16, 17, 18, 19], and we have consistently observed that the movements and sensitivities of oxalate appear to parallel those of Cl^-. The secretion of oxalate is readily activated by maneuvers that elevate cAMP, is achieved primarily by an increase in the blood-to-lumen unidirectional flux of oxalate, is inhibited by loop diuretics (bumetanide and furosemide) applied to the basolateral side [15], and is partially blocked by apical addition [18] of diphenylamine carboxylate-like compounds (DPC and NPPB). Because these characteristics of cAMP-stimulated oxalate secretion are comparable to cAMP-stimulated Cl^- secretion in the intestine [20, 21], we have proposed that oxalate shares the same (or similar) transport pathways that provide for the net electrogenic secretion of chloride [15, 18]. It is interesting, then, that in the present study there is a significant degree of dissociation between oxalate and chloride fluxes across the distal colon. This is best exemplified in Fig. 3, which depicts the induction of net oxalate secretion across the distal colon in the absence of overt net chloride secretion. Recently, we reported observing a similar phenomenon in another study [6] involving rats with two intact kidneys that were oxalate-loaded by way of an acute oxalate load (intraperitoneal injection of sodium oxalate, 3 mg/100 g body weight) or by administration of ethylene glycol (0.75%) for a 4-week period. In both of these oxalate-loaded rat models, colonic oxalate secretion was induced [6]. However, the changes in oxalate transport were not accompanied by changes in Cl^- transport, which we had expected would occur. Both in the previous [6] and present study, the dissociation of colonic oxalate and Cl^- fluxes occurred in oxalate-loaded rats having one aspect in common, namely a marked hyperoxalemia. At this time, we can only speculate on the mechanisms underlying this phenomenon, which must involve the recruitment of other transport pathways for enteric oxalate secretion in the setting of a systemic oxalate challenge. Further studies are warranted to elucidate the mechanistic basis for this dissociation in colonic oxalate and electrogenic Cl^- secretion.

It is also noteworthy that the magnitude and direction of colonic chloride transport in the CH-CRF rat is sensitive to the effects of the AT_1 antagonist, losartan (see Fig. 4), and this is entirely consistent with previous observations of the losartan sensitivity of Cl^- fluxes in studies of 5/6 nephrectomized CRF rats [5]. This result, however, is very different from the effect of losartan on colonic oxalate secretion induced in rats with normal renal function. As mentioned above, colonic oxalate secretion can be induced by 4 weeks of ethylene glycol (0.75%) treatment of rats with both kidneys intact and with normal renal function [6]. However, ANG II receptor antagonism with losartan had no significant effect on the net colonic secretion of oxalate in that rat model, which exhibited similar degrees of both hyperoxalemia and hyperoxaluria [6] as the CH-CRF model used here (and both models were generated by administering 0.75% ethylene glycol for 4 weeks). The differences in these results are significant because they indicate that multiple regulatory mechanisms (ANG II-dependent/independent, or other) are involved in mediating oxalate secretion across the mammalian distal colon. These results are also important to our understanding of adaptive enteric oxalate handling in CRF since colonic oxalate secretion in the CH-CRF rat and in the 5/6 nephrectomized rat model appear to be similarly mediated via ANG II agonism.

General Conclusions

In a previous report of renal and intestinal handling of oxalate following oxalate loading in rats [6], we concluded that (i) hyperoxaluria, per se, does not induce alterations in colonic oxalate handling and (ii) adaptive enteric oxalate secretion can be correlated with elevations in plasma oxalate in the absence of overt renal insufficiency. The latter conclusion may now be refined on the basis of the present results to indicate that some degree of renal insufficiency seems to be required to induce ANG II-mediated colonic oxalate secretion, whereas an elevation in plasma oxalate alone leads to colonic oxalate secretion that is largely independent of ANG II mediation.

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References

1. Costello JF, Smith M, Stolarski C, Sadovnic MJ (1992) Extrarenal clearance of oxalate increases with progression of renal failure in the rat. J Am Soc Nephrol 3:1098
2. Hatch M, Freel RW, Vaziri ND (1994) Intestinal excretion of oxalate in chronic renal failure. J Am Soc Nephrol 5:1339
3. Hatch M, Freel RW, Vaziri ND (1999) Regulatory aspects of oxalate secretion in enteric oxalate elimination. J Am Soc Nephrol 10:S324
4. Hatch M (1990) Spectrophotometric determination of oxalate in whole blood. Clin Chim Acta 193:199
5. Hatch M, Freel RW, Shahinfar S, Vaziri ND (1996) Effects of the specific angiotensin II receptor antagonist losartan on urate homeostasis and intestinal urate transport. J Pharmacol Exp Ther 276:187
6. Hatch M, Freel RW (2002) Renal and intestinal handling of oxalate following oxalate loading in rats. Am J Nephrol 23:18
7. Hatch M, Freel RW, Vaziri ND (1998) Losartan antagonism of angiotensin-II-induced potassium secretion across rat colon. Pflugers Arch 436:717
8. Hatch M, Freel RW, Vaziri ND (1999) AT_1 receptor up-regulation in intestine in chronic renal failure is segment specific. Pflugers Arch 437:881
9. Hoppe B, Kemper MJ, Bokenkamp A, Portale AA, Cohn RA, Langman CB (1999) Plasma calcium oxalate supersaturation in children with primary hyperoxaluria and end-stage renal failure. Kidney Int 56:268
10. Kasidas GP (1988) In: Rose GA (ed) Oxalate metabolism in relation to urinary stone. Springer, Berlin Heidelberg New York, pp 45–64
11. Mansell MA, Watts RWE (1988) In: Rose GA (ed) Oxalate metabolism in relation to urinary stone. Springer-Verlag, London, p 167
12. Greger R, Lang H, Oberleithner H, Deetjen P (1978) Handling of oxalate by the rat kidney. Pflugers Arch 374:243
13. Weinman EJ, Frankfurt SJ, Ince A, Sansom S (1978) Renal tubular transport of organic acids. J Clin Invest 61:801
14. Knight TF, Sansom S, Senekjian HO, Weinman EJ (1981) Oxalate secretion in rat proximal tubule. Am J Physiol 240:F295
15. Hatch M, Freel RW, Vaziri ND (1994) Mechanisms of oxalate absorption and secretion across the rabbit distal colon. Pflugers Arch 426:101
16. Hatch M, Freel RW, Vaziri ND (1993) Characteristics of the transport of oxalate and other ions across rabbit proximal colon. Pflugers Arch 423:206
17. Hatch M, Freel RW, Goldner AM, Earnest DL (1984) Oxalate and chloride absorption by the rabbit colon: sensitivity to metabolic and anion transport inhibitors. Gut 25:232
18. Freel RW, Hatch M, Vaziri ND (1998) Conductive pathways for chloride and oxalate in rabbit ileal brush-border membrane vesicles. Am J Physiol 275:C748
19. Hatch M, Freel RW (1995) Alterations in intestinal transport of oxalate in disease states. Scanning Microsc 9:1121
20. Barrett KE, Keely SJ (2000) Chloride secretion by the intestinal epithelium: molecular basis and regulatory aspects. Annu Rev Physiol 62:535
21. Frizzell RA, Morris AP (1994) Chloride conductances of salt-secreting epithelial cells. Curr Top Memb 42:C148