Ethylene glycol induces hyperoxaluria without metabolic acidosis in rats

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Submitted 24 January 2005; accepted in final form 20 April 2005

Green, Mike L., Marguerite Hatch, and Robert W. Freel. Ethylene glycol induces hyperoxaluria without metabolic acidosis in rats. Am J Physiol Renal Physiol 289: F536–F543, 2005. First published April 26, 2005; doi:10.1152/ajprenal.00025.2005.—Ethylene glycol (EG) consumption is commonly employed as an experimental model of hyperoxaluria, whereas metabolic acidosis is induced by EG overdose in humans. We tested the hypothesis that EG consumption (0.75% in drinking water for 4 wk) induces metabolic acidosis by comparing arterial blood gases, serum electrolytes, and urinary chemistries in five groups of rats: normal controls (CON), those made hyperoxaluric (HYP) by EG administration, unilaterally nephrectomized rats fed EG (HRF), and a reference group imbibing sweetened drinks (UNI), unilaterally nephrectomized Sprague-Dawley rats: normal controls (CON), those made hyperoxic by EG administration, unilaterally nephrectomized rats fed EG (HRF), and a reference group imbibing sweetened drinks (UNI). All rats were nephrectomized at 4 wk of age and fed a standard diet. The chemical composition of the diet is provided in Table 1. All experimental protocols were conducted in accordance with the guidelines of the University of Florida Institutional Animal Care and Use Committee and the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Materials and Methods

Animals

A total of 80 male Sprague-Dawley rats (275–300 g) were utilized in the current study and were purchased from Harlan (Indianapolis, IN). All rats had free access to Purina Rat Chow 5001 during the entire course of the study. The chemical composition of the diet is provided in Table 1. All experimental protocols were conducted in accordance with the guidelines of the University of Florida Institutional Animal Care and Use Committee and the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Experimental Models and Protocols

Two experimental models of EG-induced hyperoxaluria, together with their respective controls, were examined in the current study, which provided varying degrees of hyperoxaluria. A fifth group of rats

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Table 1. Chemical composition of the diet provided ad libitum to rats

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was made acidic and served as positive controls for metabolic acidosis.

**Control group.** For the control group (CON), 17 rats were provided free access to food and normal drinking water served as controls.

**Hyperoxaluric group.** For the hyperoxaluric group (HYP), 17 rats were given free access to food and drinking water that contained 0.75% (vol/vol) EG (changed daily) for a period of 4 wk (21). The provision of this dose of EG has generated hyperoxaluria in as early as 3 days (28) and as long as 60 days (47) with no discernable effect on renal function as judged by creatinine clearance (21, 27, 29).

**Unilateral nephrectomy group.** For the unilateral nephrectomy group (UNI), 17 rats were unilaterally nephrectomized (see below) and given 1 wk to recover. These nephrectomized rats were provided free access to food and normal drinking water served as a nephrectomy control group.

**Hyperoxaluria-induced chronic renal failure group.** For the hyperoxaluria-induced chronic renal failure group (HRF), 17 unilaterally nephrectomized rats were given 1 wk to recover from surgery before being given free access to food and drinking water that contained 0.75% (vol/vol) EG for a period of 4 wk as previously described (20).

**Metabolic acidosis group.** For the metabolic acidosis group (MA), acidosis was produced by providing free access to food and drinking water that contained 0.28 M NH₄Cl plus 5% (wt/vol) sucrose for 4 (n = 6) or 14 days (n = 6). This is a well-established protocol for the induction of metabolic acidosis in the rat (1, 38, 39). An initial analysis indicated no significant differences between the two acidic groups for any parameter examined, so the rats were combined into a single metabolic acidosis group (n = 12).

**Unilateral Nephrectomy**

To produce oxalate-induced chronic renal failure, unilateral nephrectomies were performed on 34 rats. Briefly, a surgical plane of anesthesia was induced by an intraperitoneal injection of pentobarbital sodium (40 mg/kg body wt). A small dorsal incision, ~1.5–2 cm, was made along the upper flank overlying the left kidney. The kidney was exposed through the dorsal incision, decapsulated, and the renal vasculature was ligated before excision of the renal mass. The flank ing musculature was sutured, and the skin was closed using Autoclip wound clips. Before treatments were initiated, all rats that underwent surgery were given 1 wk to recover.

**Urine Collection**

Two weeks after the initiation of treatment and on the penultimate day of the study (4 wk), rats were placed in metabolic cages and 24-h urine collections were made. Urine was collected in 20 μl of 20% sodium azide as a preservative. Particulate matter was sedimented by low-speed centrifugation. A 5-ml aliquot was removed, and the remainder was acidified by the addition of 3 N HCl (~1 ml/5 ml urine volume). The acidified urine was used for the determination of citrate, calcium, and oxalate, whereas osmolality, phosphate, chloride, titratable acid, and ammonium excretion were determined from the nonacidified aliquot.

**Blood Collection**

At the end of their respective treatment period, rats were anesthetized with an intraperitoneal injection of pentobarbital sodium (40 mg/kg body wt). Arterial blood samples (~1 ml) were drawn from the abdominal aorta (n = 11 rats/treatment group) with blood-gas sampling syringes (PROVENT®4.125, Portex, Keene, NH), and the samples were immediately transported to the STAT lab of Shands Hospital at the University of Florida for blood-gas analysis with an ABL system 500 (Radiometer, Westlake, OH). Arterial blood samples were also drawn from the acidic reference groups (MA; n = 12) and processed as described above. Base excesses were calculated with an algorithm provided by Radiometer and represent the concentration of titratable base when the blood is titrated with strong base or acid to a plasma pH of 7.4 at a PCO₂ of 40 Torr and 37°C at the actual oxygen saturation (37). A separate group of rats (n = 6 rats/treatment group) was utilized for analysis of serum electrolytes. Serum Na⁺, K⁺, and Cl⁻ and CO₂ were measured with a Roche Hitachi Modular P800 chemistry analyzer (Roche Diagnostics, Indianapolis, IN) in venous blood drawn from the anterior vena cava. Anion gaps were calculated from the serum electrolytes as follows: anion gap = serum [Na⁺] – (serum [Cl⁻] + serum [CO₃²⁻]) (43). We did not perform serum electrolyte analysis on the MA reference group as it is well established that NH₄Cl causes a hyperchloremic (normal anion gap) acidosis (4, 43). All rats were fully exsanguinated via cardiac puncture, and the serum was collected by centrifugation at 3,000 g for 15 min. An aliquot was immediately processed for oxalate determination with all precautions to prevent oxalogenesis (18), and the remainder of the serum was frozen for osmolality and creatinine determination.

**Biochemical Determinations**

Urinary chloride concentrations were determined with a chloridometer (Labcono, Kansas City, MO). Urine and serum osmolalities were measured with a freezing-point osmometer (Fiske Associates, Norwood, MA). Free orthophosphate (phosphate) was measured with a malachite green phosphate assay kit (BioAssay Systems, Hayward, CA). Creatinine was determined in the urine and serum samples using a modification of the Jaffé reaction as described by Slot (45) and further modified by Heinegard and Tiderstrom (24). Urinary ammonium was measured with an ammonium ion-selective electrode (dect/ION 3051, World Precision Instruments, Sarasota, FL). Calcium (Pointe Scientific, Lincoln Park, MI), citrate (R-Biopharm, Marshall, MI), and urinary oxalate (Trinity Biotech, St. Louis, MO) were measured with commercially available kits. Serum for oxalate determination was ultrafiltered using an Amicon Ultra-4 device, and oxalate was measured as previously described (18). Titratable acid was quantitated by titrating the urine samples to pH = 7.4 with either 1.0 N NaOH or 1.0 N HCl.

**Statistical Analyses**

All data were subjected to least-squares ANOVA using the general linear models procedures of the Statistical Analysis System (3). Significant treatment effects were separated by a Student-Newman-Keuls sequential range test (46). Main effects of EG and unilateral
nephrectomy were tested by orthogonal contrasts. When the main effect of unilateral nephrectomy was significant, comparisons were made between the UNI rats and the HRF rats. Differences between the MA group and the CON group were compared by an unpaired Student’s t-test. All data were tested for heterogeneity of variance with the Levene median test and for normality with the Kolmogorov-Smirnov test before ANOVA procedures (12). When normality and/or heterogeneity of variance tests failed, data were rank transformed before ANOVA. All data are expressed as means ± SE, and differences between means were considered statistically significant if P < 0.05.

RESULTS

Ingestion of excess non-carbonic acid loads or of compounds that are metabolized to such acids (e.g., EG and NH4Cl) may produce a metabolic acidosis characterized by a fall in arterial pH and HCO3− concentration at normal PCO2. Additionally, the anion gap [serum [Na+] − (the sum of serum [Cl−] + serum [CO2])] produced by non-carbonic acid ingestion may be elevated in metabolic acidosis.

Blood-Gas and Electrolyte Analyses

Comparisons of arterial blood-gas analyses for the five groups of rats are shown in Fig. 1. Metabolic acidosis caused a significant reduction in arterial pH (Fig. 1A; 7.43 ± 0.01 vs. 7.33 ± 0.02 in CON vs. MA, respectively) and arterial bicarbonate concentrations (Fig. 1B; 29.1 ± 0.6 vs. 21.6 ± 1.1 meq/l in CON vs. MA, respectively). However, arterial pH and bicarbonate concentrations did not statistically differ among CON, HYP, UNI, and HRF, but the lowest numerical values for each were consistently observed in HRF rats. PCO2 (Fig. 1C) and PO2 (83.8 ± 4.8 Torr in CON) were unaffected by any of the treatments (data not shown). However, there was a tendency for PCO2 to be lower in the MA group, which may represent a respiratory compensation for metabolic acidosis. Base excess was normal in the HYP group relative to the CON group (Fig. 1D). In contrast, base excess tended to be reduced in the HRF rats relative to CON rats, whereas a base deficit of 1.4 ± 1.6 meq/l was observed in the MA rats.

Serum Na+, K+, and Cl− were also measured in all rats except the MA group (Table 2). Anion gaps were calculated from these data and the serum bicarbonates (Fig. 1B). As shown in Table 2, only the unilaterally nephrectomized rats treated with EG exhibited a significant elevation in anion gap, indicating the accumulation of anions other than chloride and bicarbonate (perhaps EG metabolites like glycolate, glyoxy-late).

Thus these results demonstrate that chronic EG ingestion, at the dosages provided in this study, does not have an impact on the acid-base chemistries of rats with normal renal function (see below). The more severe model, coupling reduced renal mass with EG ingestion, appears to exhibit some, but certainly not all, of the characteristics of metabolic acidosis. In contrast, the conventional model of NH4Cl-induced metabolic acidosis exhibited the primary hallmarks of this state: decreased arterial pH with a fall in plasma bicarbonate and a base deficit.

Urine Chemistries

Metabolic acidosis may also be manifest in urinary chemistries as a reduction in urinary pH, an increase in the excretion of titratable acid (principally phosphate), and an increase in urinary ammonium ion excretion. Additionally, urinary citrate excretion, a principal organic anion of urine, is reduced in acidosis (16) and calcium excretion may be enhanced (6). Consequently, changes in urinary chemistries in the five experimental groups were evaluated using two 24-h urine collections. Collections for the CON, HYP, UNI, and HRF groups were made at 14 and 28 days and are depicted separately as noted below. In the MA group, there were no significant differences in the parameters measured in 4- and 14-day collections; hence these were combined and are depicted as noted below as a single time point (4–14).

Urinary pH after 2 and 4 wk of treatment followed a similar pattern and was not different among CON, HYP, and UNI rats, as illustrated in Fig. 2A. In contrast, urinary pH was significantly lower in the HRF rats than in the CON, HYP, and UNI rats at 2 and 4 wk, but this fall in urinary pH was not nearly as
The lack of a significant acid load in CON rats among CON, HYP, UNI, and HRF rats, but was higher in MA rats (Fig. 2). In contrast, urinary excretion of ammonium did not differ between CON and HYP rats at 2 or 4 wk. EG-induced hyperoxaluria (HYP group) had no effect on excretion of citrate in the urine after either 2 or 4 wk compared with the CON group. However, unilateral nephrectomy (UNI group) caused a modest rise in urinary citrate excretion, and this increase was attenuated in HRF rats, suggesting some tendency toward acidosis after 4 wk on EG treatment which correlates with the reduction in renal function as judged by a twofold increase in serum creatinine and a 50% reduction in renal creatinine clearance as described in a subsequent section.

Urinary excretion of calcium (Fig. 4B) was significantly increased with metabolic acidosis (MA group) and unilateral nephrectomy (UNI group) but tended to be reduced in all hyperoxaluric groups compared with their appropriate controls (HYP vs. CON and HRF vs. UNI). This trend was apparent after both 2 and 4 wk on treatment.

### Oxalate Handling

Consistent with our previous studies that utilized these rat models (20, 21), the EG-treated rats (HYP and HRF) exhibited significant hyperoxaluria, hyperoxalemia, and an increased renal clearance of oxalate compared with their respective controls (Fig. 5). By 2 wk, urinary oxalate excretion was increased about four- and sevenfold in HYP and HRF rats, respectively, compared with CON rats. Further significant increases were apparent at 28 days of EG treatment. The significant elevation in serum oxalate and the reduced renal clearance of oxalate in HRF compared with HYP, which we have previously reported (20), is confirmed here and is clearly a direct consequence of reduced renal function in these rats. This study also confirms an earlier report (20) demonstrating no differences in oxalate handling in rats with one kidney compared with healthy controls (both kidneys intact). Interestingly, we find here that metabolic acidosis is not associated with any significant alterations in oxalate homeostasis, as judged by results showing that urinary oxalate excretion, serum

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Values are means ± SE. CON, normal control; HYP, rats made hyperoxic by administration of ethylene glycol; UNI, unilaterally nephrectomized control rats; HRF, unilaterally nephrectomized rats fed ethylene glycol; MA, metabolic acidosis reference group; ND, not determined; A⁺, anions; Anion gap, serum [Na⁺] – (serum [CO₂] + serum [Cl⁻]). Kidney wet weights are presented as the weight of the single remaining kidney per kilogram body weight. Unmeasured anions in the urine were calculated as the difference between the number of milliequivalents of the measured cations (Na⁺ + K⁺ + Ca²⁺ + Mg²⁺ – NH₄⁺) and the number of milliequivalents of measured anions (Cl⁻ + phosphate) as previously described (8, 41). *P < 0.05 vs. CON. †P < 0.05 vs. UNI.
oxalate concentrations, and renal oxalate clearances are all within the normal limits for MA rats (20, 21, 23). To our knowledge, this is the first report of oxalate handling in acidotic rats.

Assessment of Renal Function

Serum creatinine (Fig. 6A) and creatinine clearance (Fig. 6B) were similar among all groups examined with the exception of the HRF group. Consistent with previous investigations of this animal model (20, 23), serum creatinine was over twofold higher in HRF rats than in all other groups and, consequently, creatinine clearance was reduced by $\frac{1}{2}$ in the HRF compared with all other groups. Total acid excretion (B) was not different among CON, HYP, UNI, and HRF rats but was higher in MA rats. *P < 0.05 vs. CON. †P < 0.05 vs. UNI.

Fig. 2. Effects of hyperoxaluria (HYP group), hyperoxaluria-induced renal failure (HRF group), and metabolic acidosis (MA group) on urine pH and total acid excretion in male Sprague-Dawley rats. After 2 and 4 wk on treatment regimes (see text for details), rats were placed in metabolic cages and 24-h urine collections were performed. Urinary pH (A) was significantly lower in HRF rats than in the CON and UNI rats at 2 and 4 wk, but this fall in urinary pH was not nearly as striking as that observed in the MA group. Total acid excretion (B) was not different among CON, HYP, UNI, and HRF rats but was higher in MA rats.

EG-induced hyperoxaluria models have been employed in numerous studies of calcium oxalate nephrolithiasis, and much of our current knowledge base in experimental hyperoxaluria and calcium oxalate kidney stone disease is based on this model (20, 21, 27, 28, 34, 35, 47). Like any experimental model, EG-induced hyperoxaluria has advantages and disadvantages. EG is inexpensive and simple to deliver in drinking water, where it is rapidly absorbed and metabolized in the liver via alcohol dehydrogenase/aldehyde dehydrogenase to glycolic acid. Glycolic acid is oxidized to glyoxylic acid, which, in turn, is further oxidized to oxalic acid by glycolate oxidase (13, 31) or lactate dehydrogenase (26), thus promoting hyperoxaluria. There has been some concern, however, that this model also initiates a metabolic acidosis that may confound the interpretation of studies using this oxalate precursor (4, 5, 11). This notion undoubtedly arises from the fact that ingestion of large doses of EG by humans or animals does, indeed, induce metabolic acidosis (25). For example, there are many reports of metabolic acidosis following ingestion of sweet-tasting anti-
freeze (primarily EG) by household pets and of humans intentionally imbibing antifreeze (15, 25). Remarkably, the proposal that EG-induced hyperoxaluria models are complicated by the presence of EG-induced metabolic acidosis has not been experimentally evaluated before the present report.

Metabolic acidosis is most simply defined as a decline in systemic pH produced primarily by a reduction in systemic bicarbonate concentrations (4). A metabolic acidosis induced by the ingestion of nonvolatile acids or acid precursors, like EG, is usually associated with an increase in the anion gap due to the presence of organic anions, principally glycolate (15, 32, 44), generated by EG metabolism. Additionally, metabolic acidosis may be associated with alterations in urine chemistry that reflect biochemical/physiological responses to the increased acid load, such as decreases in urinary pH (17, 38) and urinary citrate excretion (1, 16) and increases in urinary calcium excretion (6), urinary ammonium excretion (6, 17), and phosphate excretion (6). We have evaluated these parameters in several models to test the hypothesis that EG consumption produces acidosis at dosages commonly employed to induce hyperoxaluria (20, 21) and nephrolithiasis in rats (27, 28, 35).

**Two-Kidney Hyperoxaluria Model**

Rats consuming 0.75% EG in their drinking water did not develop any signs of metabolic acidosis after 4 wk. Thus arterial pH, bicarbonate concentrations, and anion gap of these (HYP) rats were not significantly different from two-kidney controls (CON). Urinary pH, titratable acid, and the urinary excretion of citrate, calcium, ammonium, and phosphate were similar in both groups, which further supports the conclusion that EG consumption produced no significant metabolic acidosis.

**Fig. 4.** Comparison of the renal excretions of citrate (A) and calcium (B) among CON, HYP, UNI, HRF, and MA rats. See MATERIALS AND METHODS for details of the various models. Twenty-four-hour urine collections were performed after 2 and 4 wk of treatment. Statistical differences were determined relative to CON for HYP and MA groups and relative to UNI for the HRF group. A: metabolic acidosis caused a significant reduction in urinary citrate excretion. In contrast, hyperoxaluria had no effect on citrate excretion after either 2 or 4 wk. Unilateral nephrectomy caused a modest rise in urinary citrate excretion, and this increase was attenuated in HRF rats, suggesting some tendency toward acidosis. B: unilateral nephrectomy and metabolic acidosis caused a significant increase in urinary calcium excretion. This increase was attenuated in the HRF group at 4 wk. Note that the MA group is plotted vs. the right ordinate as urinary excretion of calcium in the MA rats was ~4-fold higher than that of CON rats. *P < 0.05 vs. CON. †P < 0.05 vs. UNI.

**Fig. 5.** Serum oxalate and urinary oxalate excretion in CON, HYP, UNI, HRF, and MA rats after 4 wk on their respective treatment protocols. A: urinary excretion of oxalate was increased ~4- and 7-fold in the HYP and HRF rats, respectively, by 2 wk with further increases in oxalate excretion apparent after 4 wk of ethylene glycol treatment. B: serum concentrations of oxalate were elevated in HYP and HRF rats compared with CON rats but were similar between MA and CON rats. C: renal oxalate clearances were increased in both hyperoxaluric groups (HYP and HRF) but were similar between CON and MA groups. *P < 0.05 vs. CON.
significantly different from controls further suggests that gly-

The fact that the anion gap in HYP rats was not

plasma levels of glycolate in this time frame do not rise

because glycolate oxidase never becomes saturated; hence

sis probably never emerges in this model of hyperoxaluria

44), with metabolic acidosis ensuing (7, 36). Metabolic acido-

dependent accumulation of glycolic acid in the plasma (7, 32,

particularly when given as an oral bolus, cause the saturation-
of all other groups after 4 wk of treatment.*

P

Consequently, creatinine clearance (B) in HRF rats was approximately half that

in HYP and MA rats but was compromised in HRF rats. Serum

creatinine clearance (B) in CON, HYP, UNI, HRF, and MA rats. SeeMATE-

RIs AND METHODS for details of the various models. Renal function was

assessed as judged by serum creatinine (A) and creatinine clearance (B) in CON, HYP, UNI, HRF, and MA rats. See MATER-

ials AND METHODS for details of the various models. Renal function was

normal in HYP and MA rats but was compromised in HRF rats. Serum

that this frequently employed regimen does not produce met-

abdominal acidosis. In contrast, rats in the commonly employed

NH4Cl ingestion model of metabolic acidosis (MA group) did

exhibit all of the hallmarks of acidosis: decreased arterial pH and serum HCO3-

concentrations, together with lower urinary pH and citrate excretion but elevated urinary ammonium and phosphate excretion, which engenders increases in both titratable acid and total acid excretion.

Most likely, acidosis does not develop in the HYP rat model

because the EG is delivered at lower dose over a greater time

period compared with situations that arise in a clinical envi-

ronment where accidental or intentional ingestion of antifreeze

results in an acutely high dose. Only EG, glycolate, and oxalate

accumulate in appreciable quantities in blood and/or urine (7,

14, 44) following EG ingestion. Because glycolate oxidase (GO) is one of the rate-limiting enzymes in the metabolism of EG (14, 44), high doses of EG (>2,500 mg/kg body wt), particularly when given as an oral bolus, cause the saturation-dependent accumulation of glycolic acid in the plasma (7, 32,

44), with metabolic acidosis ensuing (7, 36). Metabolic acido-

dis probably never emerges in this model of hyperoxaluria

because glycolate oxidase never becomes saturated; hence

plasma levels of glycolate in this time frame do not rise

significantly. The fact that the anion gap in HYP rats was not

significantly different from controls further suggests that gly-

colate (or other anionic metabolites of EG) does not signifi-
cantly accumulate in this animal model.

One-Kidney Hyperoxaluria Model

Unilateral nephrectomy (UNI group) did not produce metabolic acidosis as arterial pH, serum HCO3-

concentration, and the anion gap were not significantly different from CON rats. Furthermore,

there was no significant increase in total acid excretion, reduc-
tion in urinary pH, or decreased excretion of citrate in the UNI

group, as would be anticipated in acidosis, further suggesting

that reduced renal mass per se does not lead to metabolic acidosis. (Ammonium excretion in UNI rats was depressed, which is contrary to expectations of enhanced NH4+

production in acidosis but consistent with the impaired ammonia excretion that accompanies loss of renal mass.)

In contrast, while nephrectomized rats (HRF) given 0.75%

EG in their drinking water for 4 wk did not exhibit frank metabolic acidosis, there were some signs that they may be developing an acidic state. Thus although arterial pH and HCO3-

concentrations were not significantly different from either CON or UNI controls, the HRF rats did exhibit a slightly larger anion gap and had a higher urinary phosphate excretion, a lower urinary pH, and an increase in titratable acid. Ammoni-

um excretion in HRF rats was not significantly different from

UNI rats, and, as noted above, both were actually lower than in

the CON group. It should be noted that the changes in urinary chemistry suggestive of acidosis in HRF rats are quantitatively

minor compared with the MA group.

Renal Function and Oxalate Handling in Models

Of the five models examined in this study, only nephrecto-
mized rats ingesting EG (HRF) exhibited renal failure as judged by a significant fall in creatinine clearance and a significant elevation of serum creatinine concentration. This

finding is consistent with earlier studies of the HRF model and

suggests that oxalate load imposed on nephrectomized rats is a

contributing factor in promoting renal failure (20). Indeed, this experimental model was developed to mimic oxalate-related disease states like primary hyperoxaluria with renal insuffi-
ciency caused by chronic hyperoxaluria (20). The fact that the

HRF model exhibits some characteristics suggestive of a nas-
cent metabolic acidosis is not surprising because renal failure itself causes increased anion gap metabolic acidosis (4) and metabolic acidosis has been observed in patients with primary hyperoxaluria (40).

A novel finding of this study is the observation that meta-
bolic acidosis is not associated with any significant alterations

in oxalate homeostasis. In 2001, Bushinsky et al. (6) reported

that urinary oxalate excretion was significantly reduced in

nephrectomized rats (HRF) given 0.75% NH4Cl for periods of 4–14 wk and, by way of explanation, he suggested that metabolic acidosis alters oxalate metabolism. In our study, which included urine and serum oxalate measurements, as well as an assessment of renal clearance of oxalate in Sprague-Dawley rats, we find that mean urinary excretion of oxalate in Sprague-Dawley rats, (20, 21, 23). Thus we con-

clude from this study that oxalate homeostasis is not influ-

enced by metabolic acidosis. It is, however, quiet possible that

GHS rats exhibit unusual metabolic patterns because of their
tensive inbreeding.
ACKNOWLEDGMENTS

The technical assistance of Anastasia Harris, Candl Morris, and Bonnie Murphy is greatly appreciated.

GRANTS

This work was supported by grants from the Oxalosis and Hyperoxaluria Foundation (M. L. Green) and National Institute of Diabetes and Digestive and Kidney Diseases Grant DK-56245 (M. Hatch).

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