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# Evidence for Magnesium Secretion during Phosphate Depletion in the Rat (40694)

ELLEN SACHTJEN, WOLFGANG A. MEYER,[1] AND SHAUL G. MASSRY

*Department of Medicine and the Division of Nephrology, the University of Southern California School of Medicine, Los Angeles, California 90033*

Phosphate depletion in the rat is associated with the rapid development of marked magnesuria (1, 2). This phenomenon has been attributed to a decrease in the renal tubular transport of magnesium since the magnesuria occurred in the face of falling filtered load of this ion.

However, it is also possible that magnesium secretion by the renal tubule occurs during phosphate depletion and contributes to the magnesuria. The present study was undertaken to examine this question.

*Methods.* Male Sprague–Dawley rats weighing 220–280 g and 7 to 10 weeks old were housed individually in metabolic cages. During an adaptation period of 7 days, all rats were fed control diet containing 0.44% phosphate, 0.4% sodium, 0.41% calcium, and 0.03% magnesium (I.C.N. Pharmaceutical Co., Cleveland, Ohio). The rats were then randomly allocated to the phosphate-depleted group or the control group. The phosphate-depleted group (PD) received a diet identical to the control diet except that its phosphate content was reduced to 0.03%. Control animals continued to receive the control diet *ad libitum.* The rats had free access to deionized water.

Phosphate depletion lasted for 6 to 8 days. During this period both phosphate-depleted rats and the control animals were placed in plastic restraining cages several hours each day for adaptation. The day prior to the experiment, silastic catheters were placed in the left carotid artery and the right jugular vein and a blood sample was taken from the vein for the analysis of phosphate. Only PD animals with plasma phosphates less than 6.5 mg% were used in the study. On the morning of the experiment, the rats were anesthetized with halothane (Surital, Ayerst Laboratories, New York), a bladder catheter was placed and was connected to an extension which led out through the abdominal wall. The animals were then restrained in the plastic cages and allowed to recover from anesthesia for 1 hr.

After a priming dose of 5 $\mu$Ci of [$H^3$]inulin (New England Nuclear, Boston, Mass.), a constant infusion of inulin was given in the jugular vein delivering 0.25 $\mu$Ci/min in a solution containing 2.5% dextrose, 0.45% saline, and 5 meq/liter potassium chloride. This solution was delivered by a Harvard Infusion Pump (Model 975) at 40 $\mu$l/min throughout the experiment. After a 1-hr equilibration period, three 10-min control urine collections were obtained. Blood samples of 200 $\mu$l were collected in heparinized microcapillary tubes from the carotid catheters at the midpoint of the first and third clearance periods.

Solution of magnesium chloride in 2.5% glucose were then infused at a rate of 63 $\mu$l/min in the jugular vein, utilizing a parallel syringe. The concentration of magnesium chloride was adjusted to deliver 40, 80, 120, 160, 200, and 240 $\mu$g/min for 70–100 min at each rate. After at least 45 min of equilibration at each rate of magnesium delivery, three 10-min urine collections were obtained. Blood was withdrawn at the midpoint of the first and third clearance periods.

To determine the diffusible fraction of magnesium, eight PD and seven control rats received different rates of magnesium chloride infusion and were then bled. The heparinized blood samples were immediately centrifuged. The 2–3 ml of plasma was ultrafiltered at room temperature and without control of pH through an Amicon membrane (Centriflo, 50,000 $M_r$) during 15 min of centrifugation at 1000 *g*. Magnesium concentration were determined in plasma and its ultrafiltrates.

Plasma phosphate was measured by the method of Chen *et al.* (3). Plasma and urine magnesium were determined by Perkin–Elmer atomic absorption spectrophotometer

[1] Dr. Meyer is a Fellow of the Kidney Foundation of Southern California.



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Model 503 (Perkin–Elmer Corp., Santa Ana, Calif.). [$H^3$]Inulin was counted in a Beckman liquid scintillation counter Model LS 230 (Beckman Instrument Co., Irvine, Calif.). Filtered magnesium was calculated using the diffusible fraction of plasma magnesium. The percentage diffusibility of magnesium was not different from that in control animals (76.3 ± 1.3 vs 76.6 ± 0.07%) at all levels of magnesium infusion.

*Results.* The data on the body weight, the plasma concentrations of magnesium and phosphorus and glomerular filtration rates are given in Table I. The PD rats weight (238 ± 4 g (SE)) was slightly but significantly ($P < 0.01$) less than the control animals (258 ± 7 g). The concentrations of phosphorus and magnesium in plasma were significantly lower ($P < 0.01$) in the PD rats than the controls (phosphorus: 5.6 ± 0.35 vs 8.1 ± 0.30 μg/dl; magnesium: 0.95 ± 0.05 vs 1.25 ± 0.04 meq/liter). GFR was significantly lower in PD rats than control animals both before and during all levels of magnesium infusion. The changes in plasma calcium and magnesium during magnesium infusion are given in Table II. Plasma calcium progressively fell during magnesium infusion. This is in agreement with similar observations in dogs (4).

The changes in magnesium reabsorption at all levels of filtered magnesium produced by the infusion in PD and control rats are shown in Fig. 1. With the infusion of magnesium, its filtered loads increased gradually from less than 2 to almost 14 μeq/min. In the control rats evidence for secretion was not detected despite a four- to fivefold increase in filtered magnesium up to 8 μeq/min. At higher filtered loads produced by the higher rates of magnesium infusion, only five control rats displayed slight secretion of magnesium. In contrast, in PD rats evidence for magnesium secretion was demonstrated at a filtered load of 3 μeq/min and the magnitude of the secretion increased with the rise in filtered loads to reach 4–5 μg/min at filtered loads of 6–12 μeq/min.

*Discussion.* The results of the present study show that phosphate depletion in the rat is associated with hypomagnesemia, an observation previously reported by us (2) and others (1). The data also demonstrate that significant magnesium secretion occurs in PD

TABLE I. CHANGE IN BODY WEIGHT, PLASMA LEVELS OF INORGANIC PHOSPHORUS AND MAGNESIUM, AND GLOMERULAR FILTRATION RATE IN CONTROL AND PHOSPHATE-DEPLETED RATS[a]

| | *n* | Body weight (g) | Inorganic phosphorus (mg/dl) | Magnesium (meq/liter) | GFR (ml/min) during magnesium infusion (μg/min) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | 0 | 40 | 80 | 120 | 160 | 200 | 240 |
| Control | 7 | 258 ± 7 | 8.1 ± 0.3 | 1.25 ± .04 | 2.41 ± 0.8 | 2.48 ± .09 | 2.48 ± .09 | 2.28 ± .10 | 2.19 ± .09 | 2.28 ± .07 | 2.09 ± .08 |
| PD | 8 | 238 ± 4** | 5.6 ± 0.3** | 0.95 ± .05* | 1.92 ± .11** | 2.08 ± .11* | 2.01 ± .10** | 1.88 ± .09* | 1.85 ± .08* | 1.79 ± .12** | 1.74 ± .10** |

[a] Data are presented as mean ± SE; GFR = glomerular filtration rate; PD = phosphate depletion.
* $P < 0.05$.
** $P < 0.01$.

TABLE II. CHANGE IN THE PLASMA LEVELS OF CALCIUM AND MAGNESIUM DURING MAGNESIUM INFUSION IN CONTROL AND PHOSPHATE-DEPLETED RATS[a]

| Calcium mg/dl | Before magnesium infusion | During magnesium infusion (μg/min) | | | | | |
|---|---|---|---|---|---|---|---|
| | | 40 | 80 | 120 | 160 | 200 | 240 |
| Control (*n* 7) | 9.9 ± 0.3 | 8.5 ± 0.3 | 8.0 ± 0.1 | 7.8 ± 0.2 | 7.6 ± 0.4 | 7.7 ± 0.4 | 7.3 ± 0.4 |
| PD (*n* 8) | 10.6 ± 0.55 | 9.5 ± 0.3 | 9.7 ± 0.3 | 9.5 ± 0.2 | 8.4 ± 0.3 | 8.3 ± 0.4 | 8.1 ± 0.4 |
| Magnesium meq/liter | | | | | | | |
| Control (*n* 7) | 1.25 ± 0.04 | 1.57 ± 0.13 | 2.04 ± 0.17 | 2.72 ± 0.21 | 4.03 ± 0.08 | 5.58 ± 0.47 | 7.27 ± 0.35 |
| PD (*n* 8) | 0.95 ± 0.05 | 1.04 ± 0.05 | 2.08 ± 0.19 | 3.25 ± 0.29 | 4.42 ± 0.66 | 6.10 ± 0.29 | 7.14 ± 0.32 |

[a] Data are presented as mean ± SE; PD = phosphate depletion.

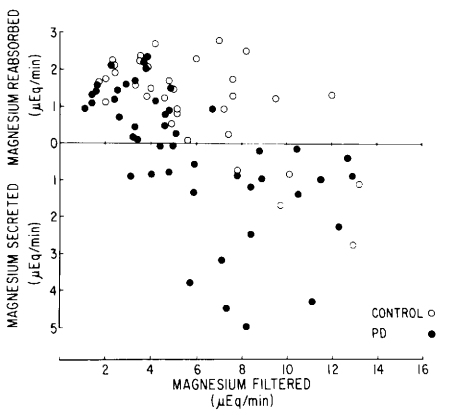


FIG. 1. The changes in the quantity of magnesium reabsorbed or secreted at various levels of filtered magnesium in control rats (○) and phosphate-depleted animals (●). Symbols below the zero line denote the magnesium secretion. Each data point is the average of two to three clearance periods obtained at the end of each level of magnesium infusion.

rats with only a twofold increase in filtered load. The magnitude of renal tubular secretion of magnesium increased as its filtered load was augmented by magnesium infusion.

In phosphate-depleted rats, the amount of magnesium reabsorbed was significantly lower and the amount secreted was significantly higher than in the control rats at any given filtered load (two-sample Hotelling $t^2$ test, $P < 0.01$). Furthermore, the slight magnesium secretion in the control animals occurred only after a four- to fivefold rise in filtered magnesium. These data clearly demonstrate that renal tubular magnesium secretion does occur during phosphate depletion and contributes to the magnesuria.

A great deal of controversy exists regarding magnesium secretion by the renal tubule. Averill and Heaton (5) have shown that the rat kidney secretes magnesium during magnesium loading. However, Alfredson and Walser (6) were unable to confirm this observation. Massry *et al.* (7) could not demonstrate magnesium secretion by the dog kidney during magnesium infusion and the superimposition of other factors known to decrease the tubular reabsorption of magnesium, such as extracellular fluid volume expansion (8), calcium infusion (9), and chronic DOCA administration (10). In contrast, Wen *et al.* (11) found that urinary magnesium exceeded filtered magnesium by 10–20% in some dogs receiving magnesium salts, saline, and furosemide. Thus, the available data suggest that if magnesium secretion by the nephron exists, it plays a minor role in the renal handling of magnesium.

Our data in normal rats also suggest that slight magnesium secretion may occur during high rates of magnesium infusion, an observation similar to that reported by Averill and Heaton (5). However, phosphate depletion appears to enhance the secretory process of magnesium by the kidney. It stands to reason, therefore, that at least part of the magnesuria of phosphate depletion (1, 2) is due to renal tubular secretion of magnesium. The mechanisms through which phosphate depletion enhances magnesium secretion are not as yet elucidated.

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