Magnesium Metabolism in Experimental Diabetes Mellitus

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SUMMARY

One of the consequences of uncontrolled diabetes mellitus (DM) is an increased urinary loss of magnesium. Some of the factors determining this electrolyte imbalance were studied in growing rats with experimentally induced DM. Two sets of experiments were performed in which dietary magnesium intake was the variable. One group each of diabetic (DB) and control (C) animals was fed a complete diet including magnesium ad libitum; the other set was given the same diet as the first two groups but the magnesium was provided daily through a stomach tube in amounts equal to those eaten by non-DB rats of the same size. All DB animals had significantly higher than normal urinary excretion of glucose, magnesium, calcium, and phosphorus. There was a positive correlation between urinary glucose and magnesium excretion. DB animals fed magnesium ad libitum became hypomagnesemic, but their bone magnesium content was increased over that of C rats. Plasma magnesium correlated negatively with both glucose and cholesterol. However, alterations in plasma cholesterol induced in C rats by dietary means had no effect on magnesium levels. When magnesium intake was restricted to the physiologic requirements of C animals, hypomagnesemia was more pronounced and occurred with concomitant depletion of magnesium in bone, suggesting possible risk of electrolyte imbalance in uncontrolled DM. The results indicate that hypomagnesemia without osseous tissue magnesium depletion may occur in experimental DM. Hyperphagia in DB animals fed ad libitum can prevent bone magnesium depletion, but a "normal" intake that does not compensate for losses may be conducive to a marked deficit in the intracellular pool of magnesium. DIABETES *26*:**882-86, September, 1977.**

Increased urinary loss of magnesium is a well-known sequela of diabetes mellitus (DM), especially when the disease is poorly controlled.[1,2] Losses of magnesium during diabetic ketoacidosis might be as high as 0.4 mmoles/kg., or about 2 per cent of body stores.[3] Similarly, in a survey of 5,100 patients with various chronic illnesses, it was shown that DM was the most frequent condition associated with hypomagnesemia.[1]

Studies of magnesium metabolism in experimental DM have demonstrated a negative magnesium balance due to increased urinary excretion and reduced net absorption of this ion.[4] Other investigators showed increased magnesium content in bone with diminished levels in skin and no alterations in other tissues.[5] An increased urinary excretion of magnesium has been reported after ingestion of glucose even in the absence of DM.[6]

However, little emphasis has been paid to alterations of mineral metabolism in DM during growth. The losses of magnesium in DM in adult organisms may have a different physiologic significance from that caused by magnesium losses or insufficient intake during growth, since the dynamics of cellular needs during development differ from those later in life.[7,8]

In this investigation, hypomagnesemia was consistently found in growing DM rats. This occurred with or without magnesium depletion. In addition, plasma magnesium levels were found to be inversely related to plasma glucose and cholesterol as a consequence of endogenous rather than dietary mechanisms.

MATERIALS AND METHODS

Male CFN rats (Carworth, NY) weighing 60 to 80 gm. were kept in a room with constant temperature of 25° C. and a 12-hour light cycle. The animals were housed in wiremesh-bottom, stainless-steel metabolic cages. After three days of acclimatization, DM was induced (day 0) by a single i.p. injection of strep-

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tozotocin (courtesy of Dr. W. E. Dulin, Upjohn Co., Kalamazoo, Michigan) at a dose 85 mg. per kilogram of body weight. Control animals received a comparable volume of saline. On day 2, 24-hour urine specimens were collected to confirm the onset of DM in rats injected with streptozotocin. On day 13 another 24-hour urine sample was obtained for determination of glucose and minerals. In addition, food and fluid intake was recorded. On day 14 all rats were anesthetized with 1.2 gm. per kilogram of i.p. urethane and exsanguinated through the abdominal aorta. The right tibia was excised, cleaned, and weighed.

Two types of experiments were performed in reference to the daily magnesium dietary intake. Diabetic and control rats were fed ad libitum a complete diet described in detail elsewhere,[7] which provided 0.51 gm. of magnesium and 5.82 gm. of calcium per kilogram of diet (diet A), or one that was magnesium-free but identical in all other aspects (diet B). Rats fed diet B were given 3.8 mg. of magnesium per 100 gm. body weight daily via gastric tube as a solution of magnesium sulfate. These requirements were derived from experiments in control rats by measuring the amount of magnesium consumed daily for two weeks. In all instances, deionized water was supplied ad libitum.

In another experiment, nondiabetic rats of similar weight were fed a complete diet A, to which cholesterol (0.5 per cent and 2.0 per cent) as well as 0.5 per cent choline chloride were added.[9] The controls received the same complete diet with no cholesterol added. The animals were killed after two weeks and blood was obtained for magnesium and cholesterol determinations.

TABLE 1

Status of diabetic and control rats two weeks after injection of streptozotocin

	Diabetic	Control
Plasma glucose (mg./dl.)	477 ± 91*	70 ± 4
Weight gain (gm./2 wk.)	8 ± 4*	33 ± 3
Urine output (ml./day)	95 ± 6*	3.3 ± 0.6
Water intake (ml./day)	111 ± 4*	5.0 ± 0.8
Food intake (gm./day/100 gm. body wt.)	15.0 ± 3.0*	9.1 ± 4.0
Magnesium intake (mg./day/100 gm. body wt.)	7.5 ± 1.5*	4.6 ± 1.9
Plasma cholesterol (mg./dl.)	103 ± 13*	54 ± 4
Arterial pH	7.30 ± 0.04	7.39 ± 0.04
Ketonemia	None	None

All rats were fed a magnesium-containing diet ad libitum. Data are given as means ± S.E.M. N = 10 to 17 rats.

*$p<0.01$ as compared with controls.

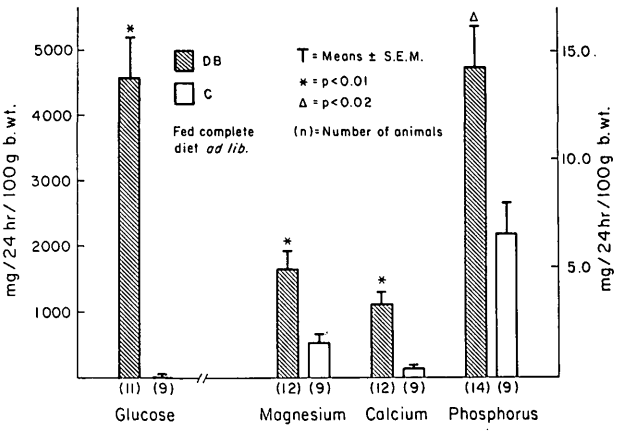

FIG. 1. Urinary excretion of glucose, magnesium, calcium, and phosphorus in rats fed ad libitum for two weeks. DB= diabetics; C= controls.

Blood samples were analyzed for glucose, magnesium, calcium, phosphorus, sodium, potassium, cholesterol, and pH. Determinations of urinary glucose, magnesium, calcium, and phosphorus were also carried out. Bone tissue was assayed for magnesium and calcium content. For diagnostic purposes urine glucose was estimated by rapid, semiquantitative methods (Labstix and Clinitest tablets). Acetest tablets were utilized for quick detection of urinary ketones. Quantitative plasma and urinary glucose was determined by a glucose oxidase method.[10] All magnesium and calcium levels were assayed by atomic absorption spectrophotometry after dilution with 0.5 per cent lanthanum chloride to suppress phosphorus interference with analysis of these ions.[11] Standard micromethods were used for the determination of cholesterol[12] and phosphorus.[13] Blood pH was measured with a blood-gas analyzer.[14]

RESULTS

Diabetic rats fed diet A gained weight at a lesser rate than the controls (table 1). They were polydipsic and polyphagic. Consequently their magnesium intake was higher than that of the controls. Diabetic animals ingested 1.6 times more magnesium than controls in relation to their weight. Other features seen in poorly controlled DM, such as polyuria, hyperglycemia, and hypercholesterolemia, were also present. Hypercholesterolemia occurred despite the lack of cholesterol in the diet. None of the animals was acidotic as judged by arterial pH. There was no ketonuria present.

Urinary excretion of glucose and minerals in diabetic animals fed diet A are presented in figure 1. All

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diabetic rats had marked glycosuria approximating 5,000 mg. of glucose per 100 gm. body weight per day. In addition, these rats had increased urinary excretion of all ions assayed as compared with controls. Magnesium losses were about 2½ times higher than those of control rats. There was also a 6½-fold increase in calcium excretion and doubling of urinary phosphorus.

The effects of experimental DM in young rats on plasma and bone magnesium are shown in figure 2. The mean plasma magnesium in animals fed diet A was 1.87 mg./dl. in the diabetic group and 2.07 mg./dl. in the control group. Despite this mild but significant hypomagnesemia, bone magnesium content in diabetic animals was about 25 per cent higher than in control rats.

Plasma magnesium in diabetic animals was correlated with the degree of hyperglycemia and hypercholesterolemia (figure 3). There was a significant negative correlation between plasma magnesium and glucose levels as well as plasma magnesium and cholesterol concentrations. Plasma magnesium was at its lowest when both plasma glucose and cholesterol concentrations were elevated. However, a similar elevation in plasma cholesterol levels induced by exogenous administration of this lipid was not associated with any significant change in plasma magnesium concentration (table 2).

The relationship between the hyperglycemia and plasma magnesium in diabetic rats seemed to be specific, since there were no alterations in other circulating electrolytes (figure 4). Diabetic animals fed diet A had no changes in plasma calcium and phosphorus levels despite the severe losses of these ions in the urine. In addition, they had no significant

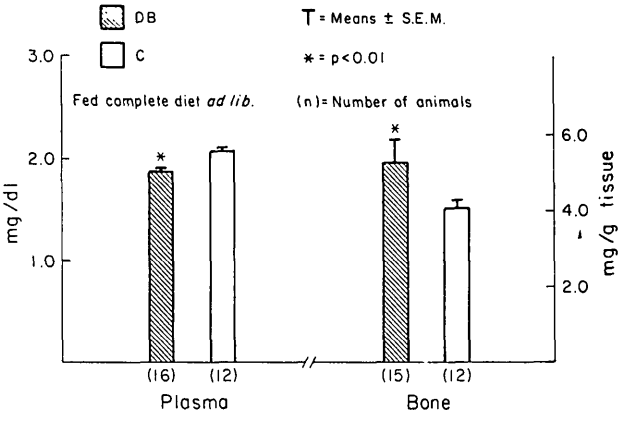

FIG. 2. Plasma and bone magnesium levels in diabetic (DB) and control (C) rats fed a complete diet ad libitum for two weeks.

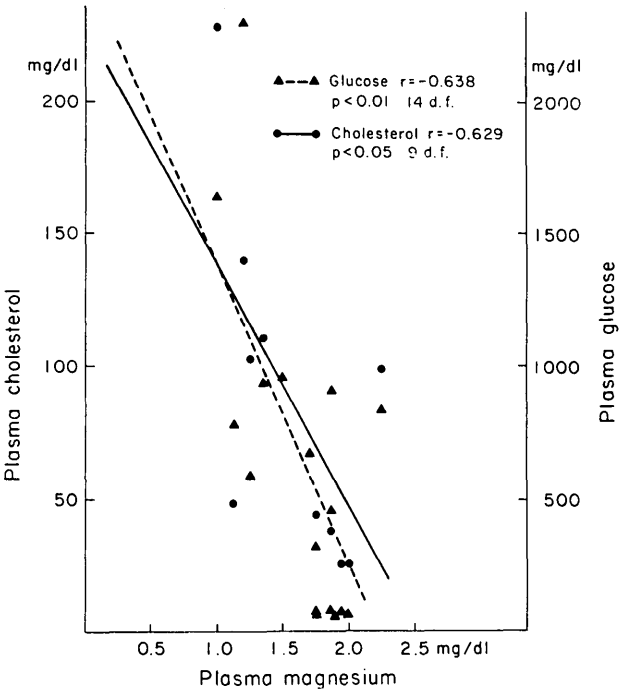

FIG. 3. Regression lines for plasma glucose and cholesterol in function of plasma magnesium in diabetic and control rats fed a complete diet ad libitum. Both negative correlations were significant at the $p<0.01$ and $p<0.05$ levels, respectively.

hyponatremia or hypokalemia.

The effects of feeding diet B in diabetic rats on plasma, bone, and urine magnesium concentrations are presented in figure 5. When the magnesium intake in diabetic rats was paired with that of control animals in proportion to body weight, diabetic rats became consistently more hypomagnesemic than those fed diet A. Moreover, they developed magnesium depletion, as suggested by low bone magnesium concentrations, that were about 60 per cent of those of controls. The urinary excretion of magnesium in diabetic

TABLE 2

Plasma magnesium and cholesterol levels in young nondiabetic rats fed complete diets with various amounts of cholesterol

	Percentage of cholesterol added		
	0%	0.5%	2.0%
Plasma cholesterol (mg./dl.)	71 ± 4 (6)	126 ± 15† (6)	180 ± 49* (7)
Plasma magnesium (mg./dl.)	2.02 ± 0.07	2.11 ± 0.04	2.02 ± 0.06

Data are presented as means ± S.E.M. (N) = number of rats in each diet group.

*$p<0.05$; †$p<0.01$ as compared with the 0 per cent cholesterol group.

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rats was higher than in control animals in spite of bone magnesium depletion and hypomagnesemia. However, the absolute amount of magnesium excreted per day was lower than in diabetic rats supplied with magnesium ad libitum.

DISCUSSION

Our principal finding was that hypomagnesemia occurs in young rats with experimentally induced DM. However, circulating magnesium levels do not reflect the nutritional status for this ion, since diabetic animals proved to have increased bone concentration of magnesium when on an unrestricted magnesium intake. This discrepancy between extracellular and intracellular distribution of this element has been previously observed.[7,8] The apparent accumulation of magnesium in the ossified tissue of diabetic animals has been reported elsewhere.[5] Conversely, substantial losses of magnesium leading to magnesium depletion can take place in the absence of hypomagnesemia.[15-17]

A negative correlation between plasma magnesium and glucose concentrations was documented. It seems unlikely that these variations were due solely to increased osmotic pressure caused by hyperglycemia, since a shift of sodium could easily compensate for glucose-induced hyperosmolality. Hyponatremia is a well-known feature characterizing inadequately controlled DM, with concomitant hyperosmolality of extracellular fluids. However, the decline in plasma magnesium was the earliest and only observable alteration in circulating electrolytes detectable during these experiments. Other ions did not show any

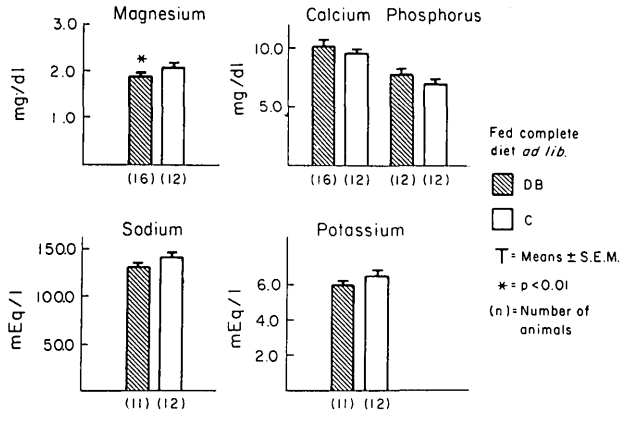

FIG. 4. Plasma electrolytes in diabetic (DB) and control (C) rats with free access to a complete diet. The only significant difference found was for plasma magnesium.

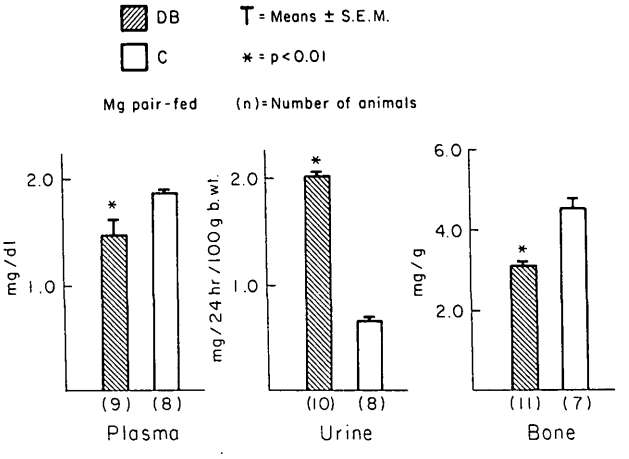

FIG. 5. Magnesium levels in plasma, urine, and bone of diabetic (DB) and control (C) rats pair-fed magnesium during the two-week experimental period. Significant declines were observed in plasma and bone levels of DB animals. These rats also lost excessive amounts of this electrolyte.

significant deviations from normal values for the duration of these studies. Therefore, it could be hypothesized that the decline in plasma magnesium levels, which is frequently seen in patients with DM,[1,2] is secondary to the hyperglycemia and/or other biochemical abnormalities of the disease and cannot be considered itself a true index of nutritional deficit.

The same negative correlation between plasma magnesium and cholesterol we observed in our studies has been reported by others.[18] In fact a decrease in hypercholesterolemia by dietary means has been attempted by providing a high magnesium intake.[19] In our experiments, plasma magnesium correlated with cholesterol levels exclusively of endogenous origin, since hypercholesterolemia in diabetic animals occurred even when a semipurified diet containing no cholesterol was given. In contrast, we were unable to demonstrate any significant plasma magnesium alterations in young, nondiabetic rats that were made hypercholesterolemic by short-term feedings of cholesterol-containing diets. This is in agreement with a recent report[20] in which an increased magnesium intake for six weeks in humans with hypercholesterolemia had no effect on plasma cholesterol. Therefore, it appears that the mechanisms relating hypercholesterolemia with hypomagnesemia in DM are independent from dietary intake and relate to the alterations in biosynthesis or catabolism of cholesterol secondary to the experimentally induced DM.

Apart from increased urinary losses of magnesium, other possible causes of hypomagnesemia and magnesium depletion seen in poorly controlled DM might be related to decreased parathyroid hormone (PTH)

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production, end-organ resistance to PTH,[21,22] inability to increased intestinal absorption of magnesium in parallel with increased intake,[4,23] and decreased heteroionic exchange of calcium for magnesium at the skeletal surface.[24]

In growing organisms, magnesium deficiency may develop not because of negative balance of this ion but as a consequence of increased demands during growth.[7,8,25,26] Therefore, the symptoms of magnesium dificiency, with or without hypomagnesemia, may have a different meaning in developing individuals than in adults. This may be particularly important in the presence of severe metabolic conditions, such as juvenile DM, and warrant a close monitoring of possible mineral deficiencies. This is an aspect of the management of juvenile DM generally overlooked in current recommendations for the medical care of DM.

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