BIOAVAILABILITY OF MAGNESIUM FROM WHEAT FLOUR AND VARIOUS ORGANIC AND INORGANIC SALTS[1]

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ABSTRACT

In weanling rats fed submarginal levels of magnesium provided by patent wheat flour, organic salts (lactate, citrate, and acetate), or inorganic salts (sulfate, oxide, chloride, phosphate, and carbonate), no outward symptoms of magnesium deficiency were observed. Serum magnesium levels were, however, subnormal. Although the absorption of magnesium from these sources did not differ significantly, magnesium concentration in various tissues examined (serum, femur, liver, and kidney) tended to be somewhat higher in rats fed wheat and magnesium-oxide and -chloride, and lower in those fed magnesium-lactate and -phosphate. This difference probably has little physiological significance, and it is concluded that magnesium was equally well available from all sources tested.

Although magnesium (Mg) has long been accepted as an essential nutrient for most animal species, data indicating its biochemical role in nutrition and possible mechanisms of its action have been presented only recently (1–5). Mg is reported to be involved in enzymatic reactions of carbohydrates, protein, and energy metabolism (1) and in the maintenance of functional and structural integrity of myocardium and other tissues (4–7). For balance in human adults, 250–400 mg of Mg per day has been suggested as necessary (8–11). On the basis of the information recently reviewed (12), a potential risk of deficiency of Mg exists among significant segments of the population. Accordingly, fortification of cereal-grain products with Mg has been suggested (12). While the significance of Mg in the diet is well defined now, little is known of the potential of various Mg sources to meet dietary needs if such a program were implemented. Work presented here was undertaken to examine the bioavailability of Mg in wheat and to compare it to various organic and inorganic sources which might be used in intended fortification. Results on the functional characteristics of these and other Mg sources in breadmaking are presented elsewhere (13).

MATERIALS AND METHODS

Weanling, male, Sprague-Dawley rats, averaging about 45 g initially were housed individually in mesh-bottom stainless-steel cages. Diets, premixed with water to minimize wastage, and deionized water were offered (eight rats per diet) *ad libitum* for 4 weeks. Body weight gain and diet intake records were kept on individual rats.

The composition of the test diets is listed in Table I. The Mg-deficient diet contained 4 mg Mg/100 g. All Mg-supplemented diets contained 19 mg of Mg/100 g, of which 15 mg was provided by wheat flour or other Mg sources. All diets contained 500 mg each of calcium (Ca) and phosphorus per 100 g—levels close to those recommended by the National Research Council (14).

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Assessment of bioavailability was based on the apparent absorption (feces collected quantitatively in the final week) and on tissue concentrations of Mg and Ca. Mg and Ca in diet, dried-pulverized feces, blood serum (blood collected by heart puncture), bone (femur), and soft tissues (liver and kidney) were determined by the method of atomic absorption spectrophotometry (15) using an IL Model 251 (Instrumentation Laboratory, Inc.) spectrophotometer. Bone ash was determined in the right femur as described earlier (16). The entire right kidney and a suitable portion of the liver were ashed overnight. The resultant ash, in all cases, was taken up in 10 ml of 6*N* HCl by heating for 15 min. Dilutions were made as necessary for the determination of Mg and Ca. Total phosphorus in the diets was determined by the standard AOAC method (17). Various Mg sources tested were (% Mg within parentheses): patent wheat flour (0.0309); sulfate (15.2); oxide (58.1); chloride (12.4); phosphate (14.0); carbonate (24.9); lactate (9.5); citrate (14.7); and acetate (11.4); determined values closely approximated formula values of Mg salts (reagent grade) used.

TABLE I
Composition of Test Diets

	Mg Source		
	None	Wheat	Others[a]
	Diet A	Diet B	Diets C–J
Casein, g	20	20	20
Vitamins[b], g	2	2	2
Alphacel[c], g	2	2	2
Trace minerals[d], g	1	1	1
Oil, g	4	4	4
NaCl, g	1	1	1
KCl, g	0.3432	0.3432	0.3432
$CaCO_3$, g	1.2381	1.2348	1.2381
$NaH_2PO_4 \cdot H_2O$, g	1.2231	1.1344	1.2231 (1.1407)[e]
Mg source, g	...	...	variable[f]
Wheat flour, g	...	61.5	...
Wheat starch, g	67.1956	5.7876	variable[g]
Calcium, mg/100 g			
Native	4.2	5.5	4.2
Added	495.8	494.5	495.8
Phosphorus, mg/100 g			
Native	225.4	245.3	225.4 (243.9)[e]
Added	274.6	254.7	274.6 (256.1)[e]
Magnesium, mg/100 g	4.0	19.0	19.0

[a]See Tables II and III.
[b]Vitamin diet fortification mixture from ICN Pharmaceuticals, Cleveland, Ohio.
[c]Nonnutritive fiber from ICN Pharmaceuticals.
[d]Contained (starch base): Mn, 5 mg; Fe, 2.5 mg; Cu, 0.5 mg; Zn, 1.2 mg; and I, 0.015 mg.
[e]Values refer to diet F (phosphate-based) only.
[f]To furnish 15 mg of magnesium.
[g]Ranged from 67.0375 to 67.1705 g to make 100 g.

RESULTS AND DISCUSSION

While requirements for a nutrient are difficult to state with accuracy, a range of 5 to 40 mg Mg/100 g of diet has been suggested as the requirement for the growing rat (3,4,7,8,14,18,19). The requirement for Mg, as for any other nutrient, is obviously dependent on a number of dietary and other factors (20–22). For example, the Mg requirement has been shown to increase with protein intake (4,5,8) and may even exceed 50 mg/100 g of diet in high-protein diets (4). Usually, dietary levels of 5 mg or less produce deficiency symptoms of varying severity in rats (7,19,23).

In the present study, Mg was provided at a submarginal level, and a high dietary protein level was provided (14), to induce mild Mg deficiency so that differences which might exist in the bioavailability of Mg from different sources could be distinguished. A submarginal level was determined based on the results of preliminary studies. After two weeks, rats fed the Mg-deficient diet (diet A) showed classical deficiency symptoms (24) characterized by erythema (ear lobes were most affected), hyperirritability, and convulsive (invariably fatal) seizures. No deficiency symptoms were detected, however, in rats fed Mg-enriched diets (diets B–J), although only about 50% of the National Research Council requirement (14) was provided (Table I). Assessment of Mg bioavailability was based on absorption, retention in various tissues, and evidence of calcinosis of soft tissues (18–21).

While no external manifestation of deficiency was observed, rats fed Mg-enriched diets showed subnormal (below 2 mg/100 ml) serum Mg levels (Table II); serum Mg levels of two surviving rats fed the deficient diet were even lower. For the maintenance of normal blood levels, the requirement for Mg, in agreement with the results of McAleese and Forbes (19), thus appears to be higher than was provided, even though it was adequate for maximum growth. Also, the availability of Mg from different sources tested differed little, since no significant difference ($P > 0.05$) in weight gain and diet efficiency was observed.

Although subnormal and not different significantly, serum Mg levels tended to be somewhat higher in rats fed diets enriched with wheat flour, Mg-oxide, or Mg-chloride than other sources. This occurred in spite of the absence of any higher absorption of dietary Mg from these sources (Table III). Apparently, Mg from wheat flour, oxide, and chloride was better retained (urinary losses were not measured) or was more favorably distributed in some tissues than others. Total diet, and therefore Mg intake, did not differ significantly ($P > 0.05$) among the Mg-enriched diets.

About 75% of the ingested Mg was absorbed (Table III), indicating that Mg was readily available from each source tested. From inorganic salts, an absorption of about 60% of ingested Mg has been reported with rats (21) and chickens (25); the difference may be due to an enhanced Mg absorption when Ca in the diet is adequate but low (26), as in present studies, or when "true" Mg availability is measured (25). In Mg-deficient rats, Mg was very poorly absorbed. Large amounts of Mg have been reported (8) to be lost in the feces despite an inadequacy of Mg in the diet.

Mg concentration in the femur was significantly higher ($P < 0.01$) in rats fed wheat flour, Mg-oxide, or Mg-chloride, in agreement with the trend noted for serum levels. Femur weight and ash content, however, did not differ significantly

TABLE II
Concentration of Magnesium and Calcium in Various Tissues
(4-Week Experiment)

	Mg Source									
	None	Wheat	Sulfate	Oxide	Chloride	Phosphate	Carbonate	Lactate	Citrate	Acetate
	Diet A	Diet B	Diet C	Diet D	Diet E	Diet F	Diet G	Diet H	Diet I	Diet J
Weight gain, g	62.0 ± 7.0	167.0 ± 8.0	160.0 ± 9.0	158.0 ± 7.0	158.0 ± 9.0	157.0 ± 4.0	160.0 ± 5.0	151.0 ± 7.0	156.0 ± 6.0	156.0 ± 5.0
Diet: gain, ratio	3.48 ± 0.47	2.02 ± 0.08	2.15 ± 0.10	2.21 0.05	2.23 ± 0.11	2.18 ± 0.04	2.22 ± 0.07	2.23 ± 0.04	2.21 ± 0.06	2.26 ± 0.08
Serum Mg, mg/100 ml	0.85 1.31	1.58 ± 0.15	1.33 ± 0.17	1.60 ± 0.23	1.58 ± 0.17	1.44 ± 0.13	1.40 ± 0.09	1.39 ± 0.11	1.28 ± 0.18	1.43 ± 0.13
Serum Ca, mg/100 ml	10.4 13.4	10.0 ± 0.5	10.0 ± 0.8	9.9 ± 0.9	10.3 ± 0.9	9.7 ± 0.6	10.0 ± 0.5	10.4 ± 0.7	10.1 ± 0.3	10.0 ± 1.1
Femur weight, mg	154.0 ± 36.0	282.0 ± 17.0	269.0 ± 34.0	259.0 ± 11.0	261.0 ± 10.0	260.0 ± 9.0	265.0 ± 17.0	254.0 ± 12.0	257.0 ± 11.0	273.0 ± 12.0
Femur ash, %	55.7 ± 2.7	59.8 ± 1.2	61.3 ± 0.8	61.6 ± 0.6	61.3 ± 0.7	60.1 ± 1.2	59.8 ± 0.5	60.2 ± 0.7	60.5 ± 0.3	60.4 ± 0.7
Femur Mg, μg	552.0 ± 74.0	1125.0 ± 124.0	985.0 ± 125.0	1092.0 ± 95.0	1125.0 ± 98.0	925.0 ± 49.0	891.0 ± 73.0	892.0 ± 92.0	829.0 ± 77.0	901.0 ± 91.0
Femur Ca, mg	43.8 ± 8.5	75.0 ± 6.7	57.5 ± 7.8	53.8 ± 2.1	54.2 ± 2.3	53.3 ± 1.3	52.2 ± 3.7	51.6 ± 2.7	53.2 ± 2.2	56.2 ± 2.1
Kidney weight, mg	188.0 ± 65.0	489.0 ± 89.0	400.0 ± 77.0	428.0 ± 60.0	469.0 ± 128.0	384.0 ± 59.0	583.0 ± 60.0	487.0 ± 63.0	503.0 ± 58.0	499.0 ± 90.0
Kidney Mg, μg	35.6 ± 7.5	121.6 ± 18.6	101.6 ± 25.0	111.1 ± 10.5	112.0 ± 25.6	85.6 ± 12.6	124.5 ± 12.9	109.0 ± 12.4	122.4 ± 13.3	120.4 ± 16.1
Kidney Ca, μg	25.7 ± 8.2	31.1 ± 4.3	43.5 ± 24.8	28.4 ± 5.1	26.8 ± 6.5	36.8 ± 6.4	51.1 ± 13.2	35.4 ± 6.1	65.6 ± 36.9	35.6 ± 5.5
Liver weight, g	1.99 ± 0.55	5.53 ± 0.56	5.16 ± 0.34	4.54 ± 0.60	4.39 ± 0.41	3.78 ± 0.41	4.91 ± 0.41	3.97 ± 0.40	4.29 ± 0.51	4.38 ± 0.43
Liver Mg, μg	482.0 ±166.0	1466.0 ± 265.0	1361.0 ± 107.0	1543.0 ± 116.0	1334.0 ± 199.0	1063.0 ± 210.0	992.0 ±161.0	974.0 ± 328.0	1147.0 ± 413.0	1326.0 ± 237.0
Liver Ca, μg	998.0 ±361.0	1478.0 ± 341.0	1348.0 ± 296.0	1345.0 ± 222.0	2073.0 ± 482.0	1918.0 ± 354.0	982.0 ±210.0	1107.0 ± 300.0	1829.0 ± 322.0	1318.0 ± 387.0

Values represent average (eight rats/diet) ± standard deviation; for diet A only, serum values for two surviving rats and averages for accumulated days rats survived.

TABLE III
Apparent Absorption of Magnesium and Calcium
(Fourth Week)

	Mg Source									
	None	Wheat	Sulfate	Oxide	Chloride	Phosphate	Carbonate	Lactate	Citrate	Acetate
	Diet A	Diet B	Diet C	Diet D	Diet E	Diet F	Diet G	Diet H	Diet I	Diet J
Mg intake, mg	1.92 ± 0.43	22.38 ± 0.93	21.10 ± 0.93	21.94 ± 1.02	21.88 ± 0.71	21.18 ± 0.81	21.91 ± 0.47	19.97 ± 1.55	21.75 ± 0.93	22.00 ± 1.19
Mg absorbed, %	36.6 ± 2.3	72.6 ± 5.8	73.1 ± 3.9	73.0 ± 4.7	73.3 ± 3.7	74.2 ± 5.2	75.1 ± 3.5	75.1 ± 5.6	73.8 ± 6.5	76.0 ± 3.7
Ca intake, mg	227.0 ± 51.0	574.0 ± 24.0	556.0 ± 24.0	563.0 ± 26.0	567.0 ± 18.0	549.0 ± 21.0	571.0 ± 12.0	520.0 ± 40.0	569.0 ± 24.0	564.0 ± 31.0
Ca absorbed, %	64.6 ± 4.8	71.3 ± 4.5	72.6 ± 1.8	71.5 ± 2.0	71.7 ± 4.9	71.7 ± 3.7	70.3 ± 2.7	72.5 ± 4.7	69.8 ± 4.1	72.3 ± 6.2

($P > 0.05$). The ash content of deficient rats was low, probably because of poorer absorption of Ca, Mg, and probably other minerals or defective calcification.

The Mg content of liver was also significantly higher ($P < 0.01$) in rats fed wheat and Mg-oxide and -chloride; those fed Mg-acetate and -sulfate also showed a higher concentration of Mg. Except for lactate- and -phosphate-fed rats, which showed consistently poorer concentrations of Mg in most tissues examined, kidney Mg levels differed little with the Mg source.

The balance technique is generally recognized as the most accurate method to determine mineral utilization (21). However, Heroux and Peter (3) recently indicated that total Mg accumulation in the carcass estimated from balance values was inconsistent with the actual content as determined by direct analysis. Presuming that the concentration of Mg in tissues sampled (Table II), rather than Mg absorption (Table III), more truly reflects the actual content of Mg in the carcass, it may be inferred that sources like wheat flour, Mg-oxide and -chloride tend to show a better bioavailability than other Mg sources. As could be expected, even when adjusted for differences in diet intake and days survived, rats fed the Mg-deficient diet showed grossly defective Mg absorption and retention.

Symptomology of Mg-deficiency varies from species to species, but calcification of soft tissues is often exhibited (23). Results in Table II show that renal and hepatic Ca content, especially when expressed as percentage of tissue weight, differed significantly ($P < 0.01$) among Mg sources tested. This may or may not be related to Mg-deficiency since, in contradiction to the results of some (19,22), others, in short-term experiments, failed to show calcinosis despite severe Mg deficiency (23,24). Essentially, the cause of soft tissue calcinosis is still unknown (22). Serum Ca levels, on enriched diets, appeared to be normal (Table II). About 70% of the dietary Ca, in agreement with the results of Cook (21) was absorbed (Table III). Serum Ca levels of the two surviving rats fed the deficient diet, however, appeared to be somewhat elevated. This is reported (24) to be true for rats, and not for most other species, including man, because of the stimulatory effect on parathyroid function and increased Ca absorption (26,27).

It may be concluded that Mg from wheat flour (not considering the possible effect of natural inhibitors such as phytates (16)) is quite well available, and that slight differences in bioavailabilities observed among other Mg sources tested may be of limited physiological significance. Unlike iron, which shows great differences in bioavailabilities depending on the source used (28), this should permit greater flexibility in considering compounds for their technical and functional aspects.

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