# Relative Bioavailability of Magnesium from Mineral- and Soy-Fortified Breads<sup>1,2</sup>

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#### **ABSTRACT**

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Effects of expanded fortification on the bioavailability of magnesium in bread were examined. Bioavailabilities were calculated using regression equations (based on serum and femur magnesium contents) developed with nonbread diets. Wheat flour provided 55% of the magnesium in test breads. The remaining magnesium (45%) was supplied by magnesium carbonate,

alone or in combination with defatted soy flour. No pattern emerged that suggests that magnesium from a single source (wheat, soy, or magnesium carbonate) is more bioavailable. This may be due, in part, to the absence of any striking differences in magnesium absorption from test breads or in fiber excretion patterns of rats.

In 1974, the Food and Nutrition Board of the National Research Council/National Academy of Sciences reviewed the current enrichment program for bread and other cereal-based foods and proposed an expansion of the program (NAS/NRC 1974). The decision to suggest increases in fortification was based on nutritional survey data that identified nutrient deficiencies. The proposed expansion included addition of 10 vitamins and minerals.

The proposed fortification can be accomplished with the use of vitamin-mineral premixes, alone or in combination with certain ingredients included in the bread formula for nutritional or functional reasons or both. Soy flour is one such ingredient (Dubois and Hoover 1981). Before the proposed fortification program is accepted, certain technical and nutritional aspects of fortification need to be studied. One nutritional aspect concerns the efficiency of utilization (bioavailability) of nutrients added to the breads. The objective of these studies was to examine bioavailability of magnesium, one of the proposed nutrients. Bread was fortified with all 10 proposed nutrients including magnesium, which originated from wheat flour, magnesium carbonate and/or soy flour.

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#### MATERIALS AND METHODS

The bread flour (11.2% protein) used in these studies was a hard red winter-spring blend that was bleached and unbromated. The soy flour was a defatted preparation (ADM Baker's Nutrisoy). The soy and wheat flours were analyzed for magnesium, calcium, zinc, and iron to calculate the amounts needed from external sources (salts) to achieve the proposed fortification levels (Table I, footnote b).

## Breadmaking

Breads were made by the sponge-dough procedure. Table II contains information on the stage, during breadmaking, when the various bread ingredients and the 10 micronutrients were added. The sponge was mixed 1 min at speed 1, using a Hobart A-120 mixer (McDuffy bowl) and fermented (31°C, 92% rh) for 3.5 hr. The dough ingredients were then mixed with the sponge for 1 min at speed 1. Mixing was continued at speed 2 until gluten developed, followed by a 20-min intermediate proof. This dough was weighed (18.5 oz), rounded, molded, placed in a bread pan, proofed (38°C, 85% rh) to template height (5/8 in.), and baked for 22 min at 224°C. Breads were air-dried at room temperature, finely ground in a food processor, and frozen in polyethylene bags until needed for analysis (Table III) and diet formulation (Table IV).

## **Bioavailability Studies**

Weanling male Sprague-Dawley rats (10 rats per diet), weighing

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<sup>&</sup>lt;sup>2</sup>Taken in part from the Ph.D. thesis by Winterringer, Kansas State University, Manhattan, 1981.

TABLE I Fortification Scheme for Test Breads<sup>a</sup>

	Minerals <sup>b</sup> (mg)											
	Iron Source			Calcium Source			Zinc Source			Magnesium Source		
Bread	Wheat Flour	Soy Flour	Fe <sup>c</sup> Salt	Wheat Flour	Soy Flour	Ca <sup>c</sup> Salt	Wheat Flour	Soy Flour	Zn <sup>c</sup> Salt	Wheat Flour	Soy Flour	Mg <sup>c</sup> Salt
A	0.83	•••		13.3		•••	0.72			24.38		•••
В-С	0.83	•••	2.80	13.3		184.9	0.72	•••	1.48	24.38	•••	19.67
D-G	0.83	0.24	2.56	13.3	7.7	177.2	0.72	0.14	1.34	24.38	7.41	12.26
H-K	0.83	0.49	2.31	13.3	15.4	169.5	0.72	0.29	1.19	24.38	14.83	4.84

<sup>&</sup>lt;sup>a</sup>Vitamins added (mg/100 g flour): vitamin A (as palmitate), 0.29; thiamin (as mononitrate), 0.64; riboflavin, 0.40; niacin, 5.29; vitamin B<sub>6</sub>, 0.44; and folic acid, 0.07 (NAS/NRC proposed levels).

TABLE II Bread Formulation (100% flour basis)

	Sponge						Dough						
Bread	Wheat <sup>a</sup> Flour	Yeast <sup>b</sup> Food	Yeast (compressed)	Soy <sup>c</sup> Flour	Water	Md	Wheat <sup>a</sup> Flour	NaCl	Sucrose	Fate	Soy <sup>c</sup> Flour	Water	Md
A	70	0.5	2.5	_	44	_	30	2	6	3	<del>-</del>	18	
В	70	0.5	2.5	_	44	+	30	2	6	3	_	18	_
Ċ	70	0.5	2.5		44	_	30	2	6	3	_	18	+
Ď	70	0.5	2.5	3	46	+	30	2	6	3	_	18	_
E	70	0.5	2.5	3	46	_	30	2	6	3	_	18	+
F	70	0.5	2.5	_	44	+	30	2	6	3	3	20	_
G	70	0.5	2.5	_	44		30	2	6	3	3	20	+
H	70	0.5	2.5	6	48	+	30	2	6	3	_	16	_
I	70	0.5	2.5	6	48	_	30	2	6	3	_	16	+
J	70	0.5	2.5		44	+	30	2	6	3	6	20	_
K	70	0.5	2.5	_	44	_	30	2	6	3	6	20	+

<sup>&</sup>lt;sup>a</sup> Bleached and unbromated.

an average of 43 g, were housed individually in stainless steel cages in a controlled environment (75° F, 50% rh, 12-hr light/dark cycle). Diets and distilled-deionized water were offered ad libitum for four weeks. Body weight gain and diet intake records were kept. Fecal matter was collected quantitatively throughout the feeding period, air-dried, weighed, and ground in a Thomas-Wiley Intermediate Mill.

Bread-based diets were formulated to contain (from breads) 15 mg of magnesium per 100-g diet; the reference diets contained four levels of magnesium supplied by magnesium carbonate (Table IV).

At the end of the four-week period, rats were anesthetized, and blood was collected by heart puncture and stored in the refrigerator overnight. Serum was recovered the next morning and stored in a freezer until analyzed for magnesium. The right femur was removed from each sacrificed rat, cleaned of adhering tissues, air-dried, ether-extracted, vacuum-dried (overnight at 70°C), cooled in a desiccator, and weighed. The dried bone was ashed in a muffle furnace overnight at 560°C and the magnesium content determined by atomic absorption spectrophotometry.

# Analytical

Minerals in wheat flour, soy flour, and mineral salts (Table I) and finely ground breads (Table III), and magnesium in serum, femur (Table V), and finely ground feces were determined by atomic absorption spectrophotometry using an IL model 251 spectrophotometer (Instrumentation Laboratories 1974). Lanthanum and strontium were used to suppress interferences in determinations. Total phosphorus in breads was determined by the standard AACC method (1976). Insoluble dietary fiber (neutral detergent fiber) in breads was determined by the recently approved

TABLE III Fiber and Minerals in Breads<sup>a</sup>

	D!-4	Minerals (mg/100 g)						
	Dietary Fiber		Phosp					
Bread <sup>b</sup>	(%)	Calcium	Total	Phytate	Magnesiun			
A	1.68	68.7	119.2	0.4	25.0			
В	1.75	229.2	108.8	37.4	44.7			
С	2.05	213.3	105.5	14.9	43.6			
D	2.30	213.4	126.9	49.3	42.0			
Ē	2.17	211.9	132.6	23.0	43.4			
F	2.29	220.7	133.4	44.6	42.4			
G	2.42	214.7	131.0	31.4	41.0			
Н	2.59	208.6	151.9	52.0	41.5			
Ī	2.31	200.9	150.7	19.8	40.7			
J	1.97	207.1	146.6	44.6	40.7			
K	2.03	213.2	148.2	31.8	41.4			

<sup>&</sup>lt;sup>a</sup> Air-dried

method of AACC (1976). Dietary fiber in fecal material was determined by a modified (using B. subtilis amylase) neutral-detergent method (Robertson and Van Soest 1977). Phytic acid phosphorus in bread and fecal material was determined by the method of Wheeler and Ferrel (1971).

# Statistical Analysis

The data were analyzed statistically using the analysis of variance with the source of variance being: stage of nutrient (minerals and

<sup>&</sup>lt;sup>b</sup>In the amount of wheat flour (100 g), soy flour (3 or 6 g), and mineral salts used in breadmaking to achieve the proposed (NAS/NRC) levels (mg/100 g flour) of: 3.63 (iron; level of 8.81 not considered), 198.20 (calcium), 2.20 (zinc), and 44.05 (magnesium).

<sup>&</sup>lt;sup>c</sup>Reagent grade salts used: FeSO<sub>4</sub> (Fe, 29.9%), CaSO<sub>4</sub>·2H<sub>2</sub>O (Ca, 23.3%), ZnO (Zn, 78.2%), and (MgCO<sub>3</sub>)<sub>4</sub>·Mg (OH)<sub>2</sub>·5H<sub>2</sub>O (Mg, 21.9%).

bContains monocalcium phosphate, starch, NaCl, ammonium sulfate, calcium sulfate, and potassium bromate (C.M. yeast food, Paniplus).

<sup>&</sup>lt;sup>c</sup>Baker's Nutrisoy (ADM Co.).

<sup>&</sup>lt;sup>d</sup> All 10 micronutrients (M) added in the amounts shown in Table 1 (footnotes a and b), with the four minerals adjusted for the amounts contributed by wheat and soy flours.

<sup>&</sup>lt;sup>e</sup>All-purpose vegetable shortening from partially hydrogenated soybean and cottonseed oils.

<sup>&</sup>lt;sup>b</sup>Same as in Table II.

TABLE IV **Composition of Test Diets** 

	Ingredients (%)									
Diet	Breada	Magnesium Carbonate	Calcium Carbonate	NaH <sub>2</sub> PO <sub>4</sub> · H <sub>2</sub> O	Sucrose	Othersb	Dietary Mg <sup>c</sup> (mg/100 g)			
Bread-based										
Α	60.00	•••	1.1458	1.4626	6.85	30.54	15.0			
В	33.53	•••	1.0568	1.6192	33.25	30.54	15.0			
C	34.39	•••	1.0656	1.6201	32.38	30.54	15.0			
D	35.73	•••	1.0583	1.5798	31.09	30.54	15.0			
E	34.58	•••	1.0571	1.5775	32.25	30.54	15.0			
F	35.40	•••	1.0536	1.5714	31.44	30.54	15.0			
G	36.57	•••	1.0526	1.5683	30.27	30.54	15.0			
Н	36.11	•••	1.0606	1.5374	30.75	30.54	15.0			
I	36.89	•••	1.0637	1.5341	29.97	30.54	15.0			
J	36.88	•••	1.0580	1.5409	29.98	30.54	15.0			
K	36.20		1.0560	1.5428	30.66	30.54	15.0			
Reference										
L	***		1.2399	1.1185	67.10	30.54	0.0			
M	***	0.0343	1.2399	1.1185	67.07	30.54	7.5			
N	•••	0.0685	1.2399	1.1185	67.03	30.54	15.0			
O		0.1027	1.2399	1.1185	67.00	30.54	22.5			

TABLE V Growth Response of Rats and Levels of Magnesium in Serum and Femura

Diet	Body Weight Gain (g)	Diet Intake (g)	Diet Gain (ratio)	Serum Mg (mg/dl)	Femur Weight (mg)	Femur Ash (%)	Femur Mg (µg/g)	Total Femur Mg (μg)	Mg (mg)
		(8)		Bread-Base		(,0)	(1.8/8)		(***8)
0 % Soy				Dieau-Dase	u				
A	192.9	395.3	2.05	1.069	355.39	54.64	2122.2	756.0	59.31
	± 9.9	± 14.8	$\pm 0.06$	± 0.15	± 16.5	± 0.45	$\pm 196.0$	±93.1	± 2.23
В	187.2	396.9	2.13	1.001	356.17	55.18	2063.8	731.2	59.56
_	±9.6	± 21.0	± 0.15	±0.11	± 17.7	±0.81	± 156.7	±71.2	± 3.14
C	184.0	391.3	2.13	1.082	354.90	54.75	1815.8	645.0	58.70
	± 8.7	± 18.2	$\pm 0.07$	$\pm 0.08$	± 17.1	$\pm 0.86$	± 154.5	± 67.1	$\pm 2.73$
3% Soy									
D	177.6	373.1	2.12	1.136	341.82	54.95	1982.7	676.8	56.41
	$\pm 10.1$	± 23.2	±0.14	±0.08	± 23.2	± 1.4	± 157.4	± 58.5	± 3.48
E	185.3	385.3	2.08	1.091	352.29	55.27	1918.2	675.5	57.81
	± 13.6	± 31.1	± 0.07	±0.12	± 20.2	±0.61	± 174.3	± 67.5	± 4.67
F	188.5	400.4	2.12	1.083	358.69	55.03	2125.7	764.0	60.00
_	± 14.2	± 27.1	±0.06	±0.16	± 22.1	± 0.55	± 112.9	± 78.9	± 4.06
G	181.1	397.2	2.19	1.032	353.38	54.79	2219.8	785.5	59.55
_	± 10.8	±21.0	± 0.09	$\pm 0.13$	± 17.6	$\pm 0.74$	$\pm 185.4$	±83.4	± 3.15
6% Soy									
Н	187.4	398.4	2.13	1.079	356.29	55.56	2236.2	797.5	59.76
	± 15.4	± 32.3	±0.09	±0.11	± 27.3	±0.61	± 148.4	±85.1	± 4.84
I	188.7	402.6	2.14	1.044	363.16	54.50	2120.4	769.8	60.40
	±11.8	± 34.2	$\pm 0.14$	±0.09	± 18.6	± 0.84	± 102.2	± 50.0	± 5.12
J	194.0	401.3	2.01	0.983	366.59	54.41	1856.6	680.0	60.20
	± 7.7	±21.8	$\pm 0.09$	± 0.08	± 24.6	±0.64	± 80.0	± 40.4	± 3.25
K	191.2	433.7	2.27	1.056	363.64	54.91	2091.6	761.0	65.05
	± 7.5	± 33.4	$\pm 0.13$	±0.11	± 17.0	± 0.65	$\pm 95.3$	± 52.5	± 5.01
				Reference	<b>;</b>				
M	137.0	353.8	2.59	0.576	328.65	53.89	1149.7	378.6	26.53
	± 7.7	$\pm 19.5$	$\pm 0.17$	$\pm 0.10$	$\pm 16.4$	$\pm 0.79$	$\pm 83.2$	$\pm 40.0$	±1.46
N	179.4	412.3	2.30	0.936	347.18	55.62	2027.5	704.2	61.84
	$\pm 4.9$	$\pm 28.0$	$\pm 0.16$	$\pm 0.12$	$\pm 16.9$	$\pm 0.63$	$\pm 143.5$	$\pm 62.0$	±4.21
O	182.3	410.1	2.25	1.405	342.60	54.50	2710.0	928.7	92.28
	$\pm 8.2$	$\pm 25.0$	$\pm 0.12$	$\pm 0.14$	$\pm 9.4$	$\pm 0.41$	$\pm 149.3$		

<sup>&</sup>lt;sup>a</sup> Mean values (10 rats per diet) ± standard deviation.

<sup>&</sup>lt;sup>a</sup> Air-dried and finely ground bread content adjusted to provide 15 mg of magnesium per 100-g diet.

<sup>b</sup> Contained (g/100 g): casein, 20.0; vitamins (vitamin diet fortification mixture from ICN Pharmaceuticals), 2.2; fiber (alphacel from ICN Pharmaceuticals), 2.0; trace minerals (Mn, 5 mg; Cu, 0.5 mg; Fe, 3.5 mg; I, 0.015 mg and Zn, 1.2 mg in sucrose base), 1; corn oil, 4.0; NaCl, 1.0; and KCl, 0.34.

<sup>&</sup>lt;sup>c</sup>Supplied by breads or Mg carbonate.

vitamins) and soy addition (SOA); soy level (SL);  $SOA \times SL$  interaction; and nonsoy breads (breads A-C). The data from all bread-based diets were included in the analysis of variance.

## RESULTS AND DISCUSSION

#### **Bread Analysis**

Fortified breads (breads B-K) contained 40.7-44.7 mg of Mg per 100 g of bread (Table III). The unfortified bread (bread A) was low in Mg (25 mg/100 g) as well as in calcium. The total phosphorus content of test breads differed less markedly but did increase with the level of soy used. Bread A was virtually free of phytate phosphorus. In fortified breads (breads B-K), the level of phytate phosphorus differed according to when the micronutrients were added; less phytate was hydrolyzed when micronutrients were added to the sponge stage as compared to the dough stage. The longer sponge fermentation (3.5 hr), compared to dough fermentation (about 1 hr), did not improve phytate hydrolysis (possible effect of altered pH, ionic strength and/or inhibition of phytase activity) or perhaps, as previously observed (Ranhotra et al 1974), some phytate reformation occurred after fermentation. The dietary fiber content of breads ranged from 1.68 to 2.59%. Soy flour, which contains about 7% dietary fiber (Erdman and Weingartner 1981), added insignificant amounts at the levels used. This amount probably changed during the baking process.

#### Test Diets

Breads provided 15 mg of Mg per 100-g diet (Table IV). Contribution from other diet ingredients was negligible, less than 2 mg of Mg. Fifteen milligrams is a submarginal level for the rat (NAS/NRC 1978). Except for Mg, test diets were complete in all other required nutrients (NAS/NRC 1978). The four reference diets contained 0.0, 7.5, 15.0, and 22.5 mg of Mg (as Mg carbonate) per 100-g diet.

### **Test Rats**

A total of 150 rats were used in the four-week experiment. Rats on the Mg-free diet (diet L) began exhibiting typical Mg-deficiency symptoms (Kruse et al 1932) by the fourth day, and all rats were dead within two weeks. No mortality was encountered among rats fed the other diets.

# Biological and Analytical Data

Biological and analytical data obtained during the four-week feeding studies are presented in Table V. On bread-based diets, body weight gain of rats ranged from 177.6 to 194.0 g, and the diet intake from 373.1 to 433.7 g. On soy bread-based diets, weight gains and diet intakes differed significantly because of SOA and SL (Table VI). On bread-based diets, femur weight ranged from 352.29 to 366.59 mg; only SL affected (increased) femur weight

significantly. On the reference diets, femur weights on diet M (Mg: 7.5 mg/100 g) were significantly lower compared to weights on the other two reference diets (Table V).

Bone ash (fat-free, moisture-free basis) in a mature bone accounts for about two-thirds of the bone mass; because of the short-term nature of the experiment, the maximum ash level achieved was only 55.56% (Table V). Neither SOA nor SL affected ash content significantly (Table VI).

Serum Mg on bread-based diets ranged from 0.983 to 1.136 mg/dl. These levels were slightly lower than we earlier observed (Ranhotra et al 1976) on diets containing 19 mg Mg/100 g but were quite comparable to values observed on diets supplying 15 mg Mg/100 g in another experiment (Ranhotra et al 1980). Only SL affected (increased) serum Mg levels significantly.

On the bread-based diets, femur Mg content ( $\mu$ g/g) ranged from 1,815.8 to 2,236.2 or (total content in  $\mu$ g) from 645.0 to 797.5. These

TABLE VI
F Values for Tissue Magnesium Levels, Magnesium
and Fiber Digestibilities and Relative Biological Values
of Magnesium in Breads

	Stage of	Soy Level 3% vs. 6%	Stage of Addition × Soy Level	Breads A vs. B vs. C
Body weight gain (g)	2.40*b	9.47*	0.49	1.89
Diet intake (g)	4.80*	12.12*	1.67	0.13
Diet: gain ratio	5.53	0.60	1.20	1.38
Serum Mg (mg/dl)	1.61	3.00*	0.98	1.46
Femur weight (mg)	1.74	6.22*	0.10	0.01
Femur ash (%)	1.84	0.94	3.60*	1.42
Femur Mg $(\mu g/g)$	5.69*	0.20	15.51*	12.87*
Total femur Mg (μg)	2.53*	3.07*	10.26*	7.46*
Mg intake (mg)	4.14*	10.70*	1.54	0.12
Mg absorbed (%)	0.18	0.0	0.79	2.12*
Fiber intake	3.34*	11.01*	3.46	13.33*
Fiber digestibility Biological value based on:	6.14*	16.03*	4.08*	3.01
Serum Mg (mg/dl)	3.53*	8.22*	0.32	1.43
Femur Mg (µg/g)	3.85*	1.83	10.23*	8.65*
Total femur Mg (µg)	1.32	0.03	9.82*	7.74*

<sup>&</sup>lt;sup>a</sup>Stage of addition of nutrients (minerals and vitamins) and soy.

TABLE VII Relative Biological Values of Magnesium in Bread

				Relative Biological Value <sup>a</sup>					
	So	urce of Magnesiu	m (%)	Serum	Femur	Total			
Bread <sup>b</sup>	Wheat Flour	Soy Flour	Magnesium Carbonate	Mg (mg/dl)	$egin{aligned} \mathbf{M}\mathbf{g} \ (\mu\mathbf{g}/\mathbf{g}) \end{aligned}$	Femur Mg (μg)	Mean <sup>c</sup>		
A	100.0	•••		$114.9 \pm 18.0$	112.9 ± 14.7	118.7 ± 19.4	115.5 ± 2.9		
В	55.4	•••	44.6	$105.3 \pm 13.9$	$108.3 \pm 13.1$	$113.3 \pm 17.3$	$109.0 \pm 4.0$		
C	55.4	•••	44.6	$117.9 \pm 11.3$	$92.1 \pm 10.4$	$97.4 \pm 11.5$	$102.5 \pm 13.6$		
D	55.4	16.8	27.8	$130.3 \pm 15.9$	$108.3 \pm 13.3$	$108.1 \pm 10.8$	$115.6 \pm 12.8$		
E	55.4	16.8	27.8	$120.9 \pm 15.8$	$101.0 \pm 12.9$	$105.2 \pm 9.0$	$109.0 \pm 10.5$		
F	55.4	16.8	27.8	$115.5 \pm 17.9$	$111.8 \pm 9.0$	$118.9 \pm 10.3$	$115.4 \pm 2.9$		
G	55.4	16.8	27.8	$109.5 \pm 17.5$	$119.3 \pm 11.9$	$124.1 \pm 13.6$	$117.6 \pm 7.4$		
Н	55.4	33.6	11.0	$115.4 \pm 20.9$	$120.1 \pm 13.0$	$126.1 \pm 11.7$	$120.5 \pm 5.4$		
I	55.4	33.6	11.0	$109.5 \pm 15.6$	$110.7 \pm 13.4$	$119.3 \pm 12.2$	$113.2 \pm 5.3$		
J	55.4	33.6	11.0	$101.8 \pm 8.3$	$92.7 \pm 8.3$	$101.9 \pm 6.4$	$98.8 \pm 4.3$		
K	55.4	33.6	11.0	$103.1 \pm 20.3$	$100.9 \pm 12.8$	$109.2 \pm 14.9$	$104.4 \pm 4.3$		

<sup>&</sup>lt;sup>a</sup> Mean values (except last column) of 10 rats per diet ± S.D. Values were calculated from regression equations developed with the "reference" diets. Biological value of Mg in magnesium carbonate = 100%.

<sup>&</sup>lt;sup>b</sup>Asterisk indicates significant difference at P < 0.05.

<sup>&</sup>lt;sup>b</sup>Same as in Table II.

<sup>&</sup>lt;sup>c</sup> Mean values by the three methods indicated ± standard deviation.

levels were significantly increased by SOA and SOA  $\times$  SL interactions. Total femur Mg was also significantly increased by SL.

#### Sources of Mg in Bread

Mg in test breads originated from as many as three different sources (Table VII). Wheat flour provided 55.4% of the Mg, except in bread A, where it provided all of the Mg. The remaining 44.6% of the Mg was provided as Mg carbonate, alone (breads B and C) or in combination with soy (breads D-K).

# Relative Biological Value (RBV) of Mg in Bread

Because of their greater sensitivity to dietary Mg levels, serum and femur Mg levels have come to be recognized as the variables of choice in calculation of RBVs (Cook 1973, Lo et al 1980, Ranhotra et al 1976). These variables (responses on the reference diets used to develop necessary equations) were used to calculate RBVs of Mg in bread-based diets.

Based on serum Mg levels, RBVs were calculated as follows:

RBV = 
$$\frac{\text{Serum Mg (mg/dl)} - 0.217 \times 100,}{0.0125 \times \text{Mg intake (mg)}}$$

where 0.217 is the Y intercept and 0.0125 is the slope of the regression line relating serum Mg level to Mg intake by rats fed the reference diets.

By this method, the RBVs of Mg in breads (Table VII) ranged from 101.8 to 130.3 (Mg in MgCO<sub>3</sub>-based reference diets = 100). Both SOA and SL significantly affected the RBVs (Table VI). In most cases, the RBVs were somewhat higher when soy and the 10 micronutrients were added to the sponge instead of the dough. RBVs on nonsoy breads did not differ significantly (Table VI).

Compared to the serum Mg levels, femur Mg levels are considered relatively more sensitive to dietary Mg levels (Cook 1973, Erdman et al 1980, Forbes et al 1979, Lo et al 1980, Ranhotra et al 1976). Based on femur Mg content ( $\mu$ g/g), RBVs were calculated as follows:

$$RBV = \frac{Femur Mg (\mu g/g) - 531.48 \times 100,}{23.78 \times Mg \text{ intake (mg)}}$$

where 531.48 is the Y intercept and 23.78 is the slope of the regression line.

By this method, RBVs of Mg in breads ranged from 92.1 to 120.1 (Mg in Mg carbonate = 100). When total femur Mg content ( $\mu$ g) was used in calculations, the equation was:

RBV = 
$$\frac{\text{Femur Mg } (\mu g) - 165.29}{8.39 \times \text{Mg intake (mg)}} \times 100,$$

where 165.29 is the Y intercept and 8.39 is the slope of the regression line. By this method, RBVs of Mg in breads ranged from 97.4 to 126.1 (Table VII).

Although the Mg intake of rats on the nonsoy breads (breads A-C) did not differ significantly (Table VI), the RBVs (based on femur Mg content) did, being highest on bread A where wheat flour provided all of the Mg. This may suggest that Mg in flour may be more "available" than Mg in Mg CO<sub>3</sub> (breads B and C contained 44.6% of the Mg as carbonate). However, such an interpretation is confounded when breads B and C are compared. The addition of Mg to the sponge (bread B), as compared to the dough (bread C), seems less detrimental, suggesting that the stage of addition of Mg may have affected RBVs more critically than did the source of Mg. More importantly, RBVs of some other breads in which Mg was supplied by soy flour and MgCO<sub>3</sub>, as well as by wheat, were even higher than the RBV on bread A (Table VII).

Of the two methods for calculating RBVs (based on femur Mg), the one that adjusts for differences in femur weight  $(\mu g/g)$  is likely to be more sensitive. By this method, the RBVs of Mg in breads (Table VII) differed significantly (were higher) due to SOA and SOA  $\times$  SL but not to SL (Table VI).

Strictly comparing the stage of soy addition, RBVs were somewhat higher when soy at the 3% level was added to the dough instead of the sponge; however, the reverse was true when breads contained 6% soy (Tables II and VII). This might suggest some interference with Mg absorption when high levels of soy are added to the dough; mean RBVs (all three methods) suggest the same (Table VII). Other than this, no pattern emerged that might suggest that Mg from any one source (wheat, soy, or MgCO<sub>3</sub>) is decidedly more bioavailable.

The apparent absorption (intake minus fecal losses) of Mg in breads ranged from 79.1 to 82.7%; on the reference diets (magnesium carbonate-based) absorption ranged from 84.0 to 87.8%. This suggests that Mg added as Mg carbonate is readily available. About one third of the ingested dietary fiber in breads was digested (28.2-41.6%), with no major differences noted between diet groups. Phytate balance studies yielded inconclusive results. Thus, the absence of striking differences in Mg absorption and in fiber and phytate excretion patterns may explain, in part, the absence of major differences in the RBVs of Mg in test breads.

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