Effect of dietary boron on mineral, estrogen, and testosterone metabolism in postmenopausal women[1]

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ABSTRACT

A study was done to examine the effects of aluminum, magnesium, and boron on major mineral metabolism in postmenopausal women. This communication describes some of the effects of dietary boron on 12 women between the ages of 48 and 82 housed in a metabolic unit. A boron supplement of 3 mg/day markedly affected several indices of mineral metabolism of seven women consuming a low-magnesium diet and five women consuming a diet adequate in magnesium; the women had consumed a conventional diet supplying about 0.25 mg boron/day for 119 days. Boron supplementation markedly reduced the urinary excretion of calcium and magnesium; the depression seemed more marked when dietary magnesium was low. Boron supplementation depressed the urinary excretion of phosphorus by the low-magnesium, but not by the adequate-magnesium, women. Boron supplementation markedly elevated the serum concentrations of 17β-estradiol and testosterone; the elevation seemed more marked when dietary magnesium was low. Neither high dietary aluminum (1000 mg/day) nor an interaction between boron and aluminum affected the variables presented. The findings suggest that supplementation of a low-boron diet with an amount of boron commonly found in diets high in fruits and vegetables induces changes in postmenopausal women consistent with the prevention of calcium loss and bone demineralization.—NIELSEN, F. H.; HUNT, C. D.; MULLEN, L. M.; HUNT, J. R. Effect of dietary boron on mineral, estrogen, and testosterone metabolism in postmenopausal women. *FASEB J.* 1: 394–397; 1987.

Key Words: boron • calcium • 17β-estradiol • testosterone • magnesium

CONCERNS ABOUT OSTEOPOROSIS, THE most common bone disorder in elderly women, have stimulated much interest in the nutrient calcium. Although most evidence reported indicates that massive intakes of calcium do not prevent bone loss in postmenopausal women (1), calcium intakes difficult to achieve through diet alone, those of up to 1500–2000 mg/day, are being recommended to women (2). These recommendations seem inappropriate because high-calcium intakes could lead to other disorders through effects on the metabolism of other nutrients. Surprisingly, these recommendations are made even though it is known that certain population groups with a low incidence of osteoporosis consume relatively low amounts of calcium (3). Thus, we decided to examine the possible effect on major mineral metabolism of some dietary substances other than the usual cholecalciferol, calcium, and fluoride. We chose to examine aluminum, magnesium, and boron. There are reports that pharmaceutical doses of aluminum elevate urinary calcium content (4). Also, Spencer et al. (4) found that subjects consuming a low-calcium diet and antacids containing aluminum salts excreted an elevated amount of calcium in the feces. In several animal species, variation in dietary magnesium alters the calcium content of urine and plasma (5). Recent studies with rats and chicks showed that boron affects major mineral metabolism and the response to high aluminum and low magnesium in the diet (6).

In the present study aluminum and magnesium treatments had no marked effects on calcium metabolism; thus, those findings will be presented elsewhere. Only the marked effects of dietary boron on selected indices of calcium metabolism in postmenopausal women will be presented in this brief communication.

METHODS

The study was done with 13 postmenopausal Caucasian women between ages 48 and 82. Two of the women were on estrogen therapy throughout the study. After medical, psychological, and nutritional evaluation established that they were in good health and emotionally suited for the study, each volunteer signed an informed consent after receiving both oral and written presentations of the nature of the research. The study protocol was approved by the Institutional Review Board of the University of North Dakota and the Human Studies Committee of the U.S. Department of Agriculture. The protocol followed the

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TABLE 1. *Effect in postmenopausal women of boron and aluminum on urinary excretion of calcium, magnesium, and phosphorus*

Dietary treatment, mg/day		Urinary excretion, g/24 h					
		Low-Mg diet[a]			Adequate-Mg diet[b]		
B	Al	Ca	Mg	P	Ca	Mg	P
0.25	0	0.117 ± 0.014	0.069 ± 0.005	0.67 ± 0.04	0.132 ± 0.038	0.111 ± 0.015	0.65 ± 0.06
0.25	1000	0.124 ± 0.018	0.071 ± 0.005	0.69 ± 0.02	0.128 ± 0.035	0.097 ± 0.009	0.73 ± 0.06
3.25	0	0.065 ± 0.006	0.050 ± 0.008	0.54 ± 0.02	0.104 ± 0.035	0.083 ± 0.012	0.67 ± 0.07
3.25	1000	0.073 ± 0.011	0.047 ± 0.005	0.59 ± 0.03	0.113 ± 0.041	0.089 ± 0.014	0.64 ± 0.06
		Analysis of variance, *P* values					
Boron		0.0004	0.0004	0.003	0.001	0.004	NS
Aluminum		NS	NS	NS	NS	NS	NS
Boron × aluminum		NS	NS	NS	NS	NS	NS

[a]Average excretion of seven women ± SEM during the last 20 days of each dietary period. [b]Average excretion of five women ± SEM during the last 20 days of each dietary period.

guidelines of the Department of Health and Human Services and the Helsinki Doctrine regarding the use of human subjects.

The women, who lived in a metabolic unit under close supervision for 167 days, were fed a 3-day menu rotation diet composed of conventional foods including beef, pork, rice, bread, and milk, but low in fruits and vegetables. To ensure adequacy, supplements were used to provide the indicated additional amounts of the following nutrients per day: potassium, 630 mg as potassium chloride; calcium, 135 mg as calcium gluconate; copper, 0.8 mg as cupric sulfate; iron, 18 mg as ferrous gluconate; cholecalciferol, 400 IU; and folic acid, 200 μg. All supplements were given at mealtimes. The kilocalorie intake of each volunteer was based on her energy needs as calculated with the Harris and Benedict equation (7) plus an additional 50% of basal energy expenditure for normal activity. To achieve the appropriate energy intake, all menu ingredients were increased or decreased proportionally in 200-kcal increments. The range of energy intakes among the subjects was 1600–2400 kcal. During the study, energy intake was adjusted to maintain body weight within 2% of admission weight. The diet contained 14% protein, 47% carbohydrate, and 39% fat. At an intake of 2000 kcal, the diet provided per day (as determined by analysis): 600 mg calcium, 870 mg phosphorus, 116 mg magnesium, 0.25 mg boron, and < 0.10 mg aluminum. The diet samples were prepared by our usual methods for elemental analysis using inductively coupled plasma emission spectrometry (8).

After an equilibration period of 23 days during which the basal low-boron diet supplemented with 200 mg of magnesium/day was fed, all women participated in four dietary periods of 24 days. These periods were: *1)* basal diet only, *2)* basal diet supplemented with 1000 mg aluminum as aluminum hydroxide/day, *3)* basal diet supplemented with 200 mg of magnesium as magnesium gluconate/day, and *4)* basal diet supplemented with 1000 mg of aluminum and 200 mg of magnesium/day. The treatments were arranged in a Latin square design and the supplements were fed in a double-blind fashion. The supplements were given in divided doses at mealtimes. Completion of these four 24-day periods and the equilibration period meant that the volunteers were fed a diet low in boron for 119 days. After completing this phase of the study, 12 women, including only 1 on estrogen therapy, participated in two additional 24-day dietary periods in which the basal diet was supplemented with 3 mg of boron as sodium borate/day in divided doses at mealtimes. Seven women, including the one on estrogen therapy, were fed: *1)* the boron basal diet only and *2)* the boron basal diet supplemented with 1000 mg of aluminum/day; thus, these seven women were fed a diet low in magnesium for the full 48 days. The other five women were fed: *1)* the boron basal diet supplemented with 200 mg of magnesium/day, and *2)* the boron basal diet supplemented with 200 mg of magnesium and 1000 mg of aluminum/day. All urine excreted daily was collected with care each morning at 8:00 AM. Urine was collected in plastic containers containing 6 ml of 3 M hydrochloric acid. Urinary calcium and magnesium were determined by using standard atomic absorption methodology (9). Urinary phosphorus was determined by using the method of Fiske and Subbarow (10). Urine samples from the last 20 days of each 24-day dietary period were used to obtain the means given in Tables 1 and 2.

Blood was drawn using standard phlebotomy techniques between 6:00 and 7:00 AM after 10 h of fasting. Serum was obtained on days 16 and 24 in each 24-day dietary period for the radioimmunoassay determination of 17β-estradiol and testosterone. The standard kits used for analyses (Radioassay System Laboratories, Carson, CA[2]) detected the total unconjugated form of testosterone and 17β-estradiol. The test for 17β-estradiol did not distinguish between the free and protein-bound states of this most biologically active form of naturally produced human estrogen.

[2]Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture, and does not imply its approval to the exclusion of other products that may also be suitable.

TABLE 2. *Changes in urinary excretion of calcium, magnesium, and phosphorus, and of serum 17β-estradiol and testosterone by dietary period*

	Dietary period[a]					
	Low boron				High boron	
Variable	Equilibration[b]	1	2	3	4	5	6
Urinary Ca, g/24 h	0.126	0.124 A[c]	0.109 AB	0.125 A	0.127 A	0.090 B	0.081 B
Urinary Mg, g/24 h	0.110	0.089 A	0.094 A	0.079 AB	0.095 A	0.074 AB	0.054 B
Urinary P, g/24 h	0.61	0.66 AB	0.61 AB	0.70 A	0.52 B	0.59 AB	0.61 AB
Serum 17β-estradiol,[d] pg/ml	23.9	11.9 A	15.0 A	26.9 AB	12.7 A	35.9 B	37.5 B
Serum testosterone,[d] ng/ml	0.60	0.34 A	0.31 A	0.33 A	0.30 A	0.71 B	0.64 B

[a] After an equilibration period of 23 days when dietary boron was 0.25 mg/day there were six 24-day dietary periods. In the first four periods, magnesium and aluminum were varied using a Latin square design, and boron was kept at 0.25 mg/day. In the last two dietary periods, boron was increased to 3.25 mg/day. In these two periods, seven women were not supplemented with magnesium, five women were fed a supplemental 200 mg of magnesium/day, and dietary aluminum varied. The six dietary periods are listed sequentially according to time and without regard to different dietary treatments. See text for full details. [b] Urinary excretion values represent the mean of 13 women during days 7–9 of the equilibration period, serum 17β-estradiol and testosterone values represent the mean of 12 women on day 23 of the equilibration period. [c] Values above the same letters are not significantly different ($P > 0.05$) as determined by Scheffé contrasts (10). The equilibration period was not included in the statistical analysis. [d] Means do not include values from one woman who was treated with estrogen.

Statistical treatment of the data was done by using repeated measures of analysis of variance (11). Scheffé contrasts were used to determine the significance of the differences between means (11).

RESULTS AND DISCUSSION

The data in Table 1 indicate that dietary boron had a marked effect on major mineral metabolism in the postmenopausal women. This effect apparently was modified by magnesium status, but not by the ingestion of 1000 mg of aluminum/day. Basically, the data show that supplements of 3.0 mg of boron/day fed to postmenopausal women, who had been consuming 0.25 mg of boron/day for 119 days, markedly reduced their urinary excretion of calcium and magnesium. Although the experimental design prevented a direct examination of the effect of magnesium status on the response to dietary boron, the differences in excretion seemed to be more marked in the low-magnesium than in the adequate-magnesium women. For example, the reduction of urinary calcium excretion caused by boron supplementation was 52 mg/day in the low-magnesium women and 22 mg/day in the adequate-magnesium women. Urinary phosphorus excretion was reduced by boron supplementation in the low-magnesium women but not in the adequate-magnesium women.

The data in Table 3, which does not include the woman on estrogen therapy, show that a supplement of 3.0 mg of boron/day fed to postmenopausal women who had been consuming 0.25 mg boron/day for 119 days markedly elevated the serum concentration of 17β-estradiol and testosterone. Similar to the changes in urinary excretion of minerals, the elevation in serum steroids seemed more marked in the low-magnesium women. Although it was not significant, aluminum supplementation tended to reduce the steroid response of the adequate-magnesium women to boron supplementation.

TABLE 3. *Effect in postmenopausal women of boron and aluminum on serum concentrations of 17β-*estradiol and testosterone

Dietary treatment, mg/day		Low-Mg diet		Adequate-Mg diet	
B	Al	17β-Estradiol,[a] pg/ml	Testosterone,[a] ng/ml	17β-Estradiol,[b] pg/ml	Testosterone,[b] ng/ml
0.25	0	21.1 ± 6.5	0.31 ± 0.06	15.5 ± 5.4	0.38 ± 0.09
0.25	1000	17.8 ± 4.2	0.34 ± 0.06	24.6 ± 7.9	0.33 ± 0.05
3.25	0	41.4 ± 12.1	0.83 ± 0.09	38.0 ± 1.5	0.65 ± 0.05
3.25	1000	38.5 ± 5.9	0.66 ± 0.10	29.9 ± 2.5	0.56 ± 0.05
		Analysis of variance, P values			
Boron		0.01	0.0008	0.03	0.02
Aluminum		NS	NS	NS	NS
Boron × aluminum		NS	NS	NS	NS

[a] Average serum concentration of six women ± SEM on days 16 and 24 of each dietary period. Means do not include values from one woman who was treated with estrogen. [b] Average serum concentration of five women ± SEM on days 16 and 24 of each dietary period.

The data in Table 2 indicate that the findings attributed to boron were not spurious effects caused by the passage of time under the described environmental conditions. As the experiment progressed, there was no gradual decrease in the urinary excretion of calcium and magnesium, nor a gradual increase in the serum concentration of 17β-estradiol and testosterone. All of these variables changed abruptly, regardless of dietary aluminum and magnesium, about 8 days after boron supplementation began. The equilibration values in Table 2 suggest that low-dietary boron decreases serum 17β-estradiol and testosterone; however, this must be confirmed by further experimentation.

The effect of boron on mineral metabolism most likely involved some biochemical or endocrine mechanism. It seems unlikely that urinary mineral excretion was reduced through chemical or complexing actions of boron, preventing the absorption of calcium, magnesium, and phosphorus because of the quantity of boron in the diet (3.25 mg) relative to that of calcium (600 mg), phosphorus (870 mg), and magnesium (116–316 mg).

The 17β-estradiol and testosterone findings suggest that boron prevented the urinary loss of calcium, magnesium, and phosphorus through endocrine mechanisms.

A recent review (6) of the boron content of foods indicates that those of plant origin are rich sources of boron. Meat and fish apparently are poor sources of boron. Thus, the daily intake of boron by humans can vary widely depending on the proportions of various food groups in the diet. Diets containing a variety of foods including fruits and vegetables should supply about 1.5–3.0 mg of boron/day. Although the findings need clarification by further experimentation, they strongly suggest that supplementation with boron in amounts commonly found in diets high in fruits and vegetables induces changes in postmenopausal women consistent with the prevention of calcium loss and bone demineralization. Boron may be an important nutritional factor determining the incidence of osteoporosis. FJ

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