# The justification for providing dietary guidance for the nutritional intake of Boron

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# The Justification for Providing Dietary Guidance for the Nutritional Intake of Boron

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## ABSTRACT

Because a biochemical function has not been defined for boron (B), its nutritional essentiality has not been firmly established. Nonetheless, dietary guidance should be formulated for B, because it has demonstrated beneficial, if not essential, effects in both animals and humans. Intakes of B commonly found with diets abundant in fruits, vegetables, legumes, pulses, and nuts have effects construed to be beneficial in macromineral, energy, nitrogen, and reactive oxygen metabolism, in addition to enhancing the response to estrogen therapy and improving psychomotor skills and cognitive processes of attention and memory. Perhaps the best-documented beneficial effect of B is on calcium (Ca) metabolism or utilization, and thus, bone calcification and maintenance. The paradigm emerging for the provision of dietary guidance that includes consideration of the total health effects of a nutrient, not just the prevention of a deficiency disease, has resulted in dietary guidance for chromium (Cr) and fluoride; both of these elements have beneficial effects in humans, but neither has a defined biochemical function. Knowledge of B nutritional effects in humans equals or is superior to that of Cr and fluoride; thus, establishing a dietary reference intake for B is justified. An analysis of both human and animal data suggests that an acceptable safe range of population mean intakes of B for adults could well be 1–13 mg/d. Recent findings indicate that a significant number of people do not consistently consume more than 1 mg B/d; this suggests that B could be a practical nutritional or clinical concern.

**Index Entries:** Boron; calcium; bone; dietary reference intakes; human nutrition.

# INTRODUCTION

In 1940, essential nutrients included mineral elements whose dietary deficiency led to disease or failure to grow. The use of the animal growth model to identify essential nutrients and to quantify requirements was the foundation of early experimental nutrition. In the 1960s and 1970s, the criteria for establishing essentiality of mineral elements that could not be fed in low enough amounts to cause death or the interruption of the life cycle usually included the following *(1–5)*:

1. The element must react with biological material or form chelates.
2. The element must be ubiquitous in seawater and earth's crust.
3. The element must be present in significant quantity in animals.
4. The element should be toxic to animals only at relatively high intakes in comparison to nutritional intakes.
5. Homeostatic mechanisms must exist for the element so that it is maintained in the body in a rather consistent amount during short-term variations in intake.
6. Finally, and most importantly, a dietary deficiency must consistently result in a reduction of a biological function from optimal that is preventable or reversible by physiological amounts of the element.

In the 1980s and 1990s, the use of the above criteria for establishing essentiality of mineral elements lost its general acceptance when a large number of elements were suggested to be essential because of some small change in a physiological or biochemical variable (Nielsen, personal observations). It was thought that many of these changes were not necessarily the result of a suboptimal function, but perhaps a consequence of a toxicologic or pharmacologic action. In this case, pharmacologic means the ability of a dietary intake of a substance to alleviate a condition other than the nutritional deficiency of that substance, or to alter a biochemical function or biological structure in a therapeutic manner. As a result of this change in acceptance, if the lack of an element cannot be shown to cause abnormal development leading to serious impairment in quality of life, or to interrupt the life cycle (death or interferes with growth, development, or maturation such that procreation is prevented), general acceptance of an element as essential now seems to require that it have a defined biochemical function. As a result, setting dietary guidance for elements that have no defined function and, thus, uncertain essentiality status, has become a contentious issue *(6)*. Nonetheless, this is an issue that needs to be closely evaluated for several nutrients, because the outcome could have health and economic consequences.

Boron (B) is a food component that needs consideration for dietary guidance. There is both precedence and a new paradigm for nutri-

tional guidance to support such consideration. Moreover, the circumstantial evidence for essentiality of B is so strong that for cautionary reasons, it seems appropriate to give dietary guidance for this element with regard to health and well-being until a biochemical function can be defined.

## JUSTIFICATION FOR DIETARY GUIDANCE

The provision of dietary guidance for Cr and fluorine as fluoride acts as a precedent for doing so for B *(7)*. Unequivocal or specific signs of fluoride deficiency have not been described for higher animals, including humans; thus, fluoride is not generally considered an essential element. Nonetheless, there are intakes of dietary fluoride that have beneficial actions in humans; these actions include the prevention of dental caries and possibly osteoporosis *(8)*.

As a result, an estimated safe and adequate daily dietary intake (ESADDI) for fluoride was established in 1989 *(7)*, and dietary reference intakes (DRIs) were recently provided for fluoride *(9)*. A biochemical function has not been defined for Cr, but it has been shown to alleviate some cases of impaired glucose tolerance and, in relatively high amounts, diabetes *(10–12)*. Because of its beneficial and possibly essential effects in humans, an ESADDI was established for Cr *(7)*. Like Cr and fluoride, B has been shown to have beneficial effects in humans. Thus, if the basis for providing dietary guidance for a mineral element is kept consistent, B should be treated the same as fluoride and Cr in the report of the committee establishing DRIs for trace elements.

The new paradigm emerging for the provision of dietary guidance is that the prevention of a deficiency disease or change in a biochemical function in determination of nutritional requirements is complemented by the concern for the total health effects of a nutrient *(13)*. Today, because of apparent beneficial health effects, dietary intake recommendations for nutrients are being made that far exceed those required to prevent deficiency pathology. Ca can be used to illustrate this new paradigm. Numerous studies have shown that a Ca intake of 600–800 mg/d is adequate to maintain Ca balance, and to prevent undesirable changes in indicators of Ca status or metabolism in humans *(14)*, especially when they consume diets adequate in all nutrients known to facilitate Ca utilization, including B, copper (Cu), magnesium (Mg), manganese, and vitamin D. Nonetheless, various work groups, consensus statements, and Ca metabolism authorities have vigorously proposed recommendations higher than the recommended dietary allowance (RDA) given in 1989 *(7)*, because such intakes have been shown to be of short-term benefit in preventing bone Ca loss, which can lead to osteoporosis. In other words, pharmacologic amounts of Ca, or amounts that would be difficult for many

people to achieve by diet alone, are being recommended to overcome the poor utilization of Ca caused by the lack of other needed nutrients leading to the loss of bone with aging. A result of these recommendation was DRIs *(9)* higher than the previous RDAs *(7)*. Recommended intakes higher than that necessary to prevent deficiency pathology are also being advocated for other nutrients, including folic acid, vitamin C, and vitamin E. Recommendations for intakes higher than the RDA for selenium most likely will be considered, because such intakes have been shown to protect against certain types of cancer *(15)*. Furthermore, luxuriant intakes of nonessential food components, such as antioxidants from plants and fiber that have apparent beneficial effects, are being recommended.

The emerging new paradigm attests to the judgment that a prudent action would be to provide dietary guidance for B because of its demonstrated beneficial, if not essential, effects in both humans and animals. In other words, the judgment that B should not receive much consideration by groups giving dietary guidance because of the lack of a defined biochemical function that could be used to assess the presence of deficiency pathology needs to be discarded.

## BENEFICIAL/ESSENTIAL EFFECTS OF B

The beneficial effects of B alluded to in the preceding paragraph have been found with intakes of B commonly found with diets abundant in fruits, vegetables, legumes, pulses, and nuts. B supplementation, usually as sodium borate, at those amounts after an appropriate period of B deprivation has resulted in changes in humans that can be construed to be beneficial in macromineral, energy, nitrogen, and reactive oxygen metabolism *(16)*. Furthermore, the supplementation enhanced the response to estrogen therapy *(16)*, and improved psychomotor skills and cognitive processes of attention and memory *(17)*. Moreover, numerous findings have been described that show chicks and rats fed low dietary B, or <0.3 μg/g diet, exhibit altered bone development, brain function, macromineral metabolism, energy substrate utilization, immune function, and insulin secretion *(16–18; also see* articles by Hunt and Penland in this vol.).

Because other articles in this volume present many of the findings indicating B has a beneficial effect in higher animals, including humans, only the apparent beneficial effects on Ca metabolism or utilization, and thus bone calcification and maintenance, will be presented here to support the position that nutritional guidance should be given for B.

### Animal Findings

One of the first findings reported in 1981 *(19)* suggesting that B is essential was that B improved bone calcification in chicks fed a diet deficient, but not completely lacking in vitamin D. The marginal intake of vitamin D is important because subsequent research has found that B does

not replace vitamin D, but instead enhances its utilization *(17)*. In the 1981 report, it was stated that rachitic long bones were found in 17 of 21 B-deprived, vitamin D-deficient chicks; only 9 of 22 B-supplemented, vitamin D-deficient chicks exhibited rachitic bones.

Furthermore, the degree of subnormal calcification was much more severe in the B-deprived than in B-supplemented chicks. Several subsequent reports show that B in physiological amounts enhances the utilization of vitamin D in the formation and calcification of bone in experimental animals. Hunt *(20)* showed at the microscopic level that B deprivation exacerbates the distortion of marrow sprouts caused by vitamin D deficiency, and delays the initiation of cartilage calcification. Hunt and coworkers also found that B deprivation decreased chondrocyte density in the zone of proliferation of the growth plate in vitamin D-deficient chicks *(21)*. King and coworkers *(22)* showed that *in ovo* injections of B reduced the abnormal height of long bone growth plate in chicks hatched from vitamin D-deficient chicks.

In addition to the bone findings, there are a number of other findings from animal experiments suggesting that B has a beneficial effect on Ca or macromineral metabolism. Hegsted and coworkers *(23)* found that B deprivation decreased the apparent absorption and balance of Ca, Mg, and phosphorus (P) in the vitamin D-deficient rat. In the vitamin D-deficient chick, B deprivation has been found to decrease plasma 25-hydroxycholecalciferol *(24)* and 1,25-hydroxycholecalciferol concentrations *(25)*. Regardless of vitamin D status, Hunt and coworkers *(21)* found that B deprivation depressed femur Ca, P, and Mg concentrations in chicks.

The data presented in Tables 1 and 2 (Nielsen, unpublished) are another indication that the metabolism of Ca is related to B status. The data come from a factorially arranged experiment in which male Sprague-Dawley rats were assigned to groups of 12, and fed a casein-ground corn–corn oil basal diet containing about 0.12 mg B and 3.6 g of potassium (K)/kg for 12 wk. The dietary variables were supplemental B at 0 and 3 mg/kg and K at basal 3.6 and supplemented to give 10 g/kg. The data show that whenever a dietary treatment decreased the B concentration in a tissue, it decreased the Ca concentration in that tissue. On the other hand, if a dietary treatment did not affect the B concentration of a tissue, it did not affect the Ca concentration of that tissue. For example, B deprivation depressed the B concentration and also depressed the Ca concentration in the femur. Moreover, feeding supplemental K increased both the B and Ca concentrations in femur. The end result was that the highest B concentration occurred in femurs with the highest concentration of Ca; the lowest concentration occurred in femurs with the lowest concentration of Ca. B deprivation did not decrease the concentration of B in the liver and muscle; neither did it decrease the conentration of Ca. On the other hand, feeding elevated dietary K decreased the B concentration in liver and muscle; this decrease was paralleled by a decrease in the Ca concentration.

Table 1
Effect of Dietary B and K on B and Ca Concentrations
on a Dry-Weight Basis in Brain and Femur of Rats

| Treatment | | Brain | | Femur | |
| --- | --- | --- | --- | --- | --- |
| B[2] | K[2] | B, ug/g | Ca, ug/g | B, ug/g | Ca, mg/g |
| 0 | 0.36 | 0.245±0.086 | 245±91 | 0.213±0.037 | 205±4 |
| 0 | 1.00 | 0.271±0.057 | 231±55 | 0.454±0.089 | 206±5 |
| 3 | 0.36 | 0.379±0.113 | 281±129 | 0.563±0.095 | 208±5 |
| 3 | 1.00 | 0.327±0.111 | 313±170 | 0.813±0.126 | 214±5 |
| Analysis of Variance - P Values | | | | | |
| Boron | | 0.003 | 0.10 | 0.0001 | 0.001 |
| Potassium | | 0.67 | 0.80 | 0.0001 | 0.05 |
| B x K | | 0.19 | 0.53 | 0.86 | 0.16 |

[1]Values represent the mean ± SD of 12 rats.

[2]Amounts of boron ($H_3BO_3$) in mg/kg and potassium (KCl) in percent supplemented to the diet.

## HUMAN FINDINGS

The most compelling findings indicating that B is of nutritional importance for Ca metabolism have come from two experiments in which men over the age of 45, postmenopausal women, and postmenopausal women on estrogen therapy were fed a low B diet or about 0.25 mg/2000 kcal for 63 d, and then fed the same diet supplemented with 3.0 mg of B/d for 49 d (26,27). The major differences between the two experiments were the intakes of Cu and Mg; in one experiment, Cu was marginal or about 1.6 mg Cu/2000 kcal, and Mg was inadequate or about 115 mg Mg/2000 kcal. In the other experiment, both were adequate, or about 2.4 mg Cu and 300 mg Mg/2000 kcal. Table 3 shows that B affects Ca or macromineral metabolism in humans, through changing hormones that are involved in this metabolism.

In both experiments, the serum 25-hydroxycholecalciferol concentration was lower during B depletion than B repletion (26,27). In the experiment where dietary Cu was marginal and Mg was inadequate,

Table 2
Effect of Dietary B and K on B and Ca Concentrations
on a Dry-Weight Basis in Liver and Muscle of Rats[1]

| Treatment | | Liver | | Leg Muscle | |
|---|---|---|---|---|---|
| B[2] | K[2] | B, $\mu$g/g | Ca, $\mu$g/g | B, $\mu$g/g | Ca, $\mu$g/g |
| 0 | 0.36 | 0.407±0.108 | 105±11 | 0.252±0.048 | 177±11 |
| 0 | 1.00 | 0.365±0.053 | 89±8 | 0.194±0.058 | 165±9 |
| 3 | 0.36 | 0.428±0.064 | 100±9 | 0.219±0.041 | 174±9 |
| 3 | 1.00 | 0.358±0.067 | 91±5 | 0.188±0.038 | 167±18 |
| Analysis of Variance - P Values | | | | | |
| Boron | | 0.76 | 0.63 | 0.17 | 0.88 |
| Potassium | | 0.02 | 0.0001 | 0.002 | 0.01 |
| B x K | | 0.54 | 0.12 | 0.32 | 0.49 |

[1]Values represent the mean ± SD of 12 rats.

[2]Amounts of boron ($H_3BO_3$) in mg/kg and potassium (KCl) in percent supplemented to the diet.

calcitonin values were much higher than in the experiment where these two elements were adequate. This finding suggests that because the calcitonin values obtained with adequate Cu and Mg were close to those reported by others as being normal *(28)*, the combined Mg-low, Cu-marginal diet caused elevated serum calcitonin indicative of an abnormal calcitonin metabolism. Interestingly, calcitonin has been found to be elevated in postmenopausal women with osteoporosis *(28)*. As shown in Table 3, B depletion exacerbated this abnormality. Findings similar to calcitonin were obtained with serum osteocalcin. That is, serum osteocalcin concentrations were higher in the experiment where Cu was marginal and Mg was inadequate. B depletion exacerbated this elevation. Plasma concentrations of osteocalcin have been inversely related to bone mineral content in postmenopausal women *(29)*. Finally, in both experiments, estrogen ingestion elevated serum 17$\beta$-estradiol; this elevation was higher during B repletion than B depletion (Table 3). This finding indicates that B can enhance the effects of estrogen therapy which is used to prevent bone loss in postmenopausal women.

Table 3
Effect of Dietary B on Serum Calcitonin, Osteocalcin,
and 17β-estradiol in Men, Postmenopausal Women,
and Postmenopausal Women on Estrogen Therapy

| Dietary Boron[1], | Calcitonin, pg/ml | | Osteocalcin, ng/ml | | 17B-estradiol, pg/ml | |
|---|---|---|---|---|---|---|
| mg/day | Exp. 1[2] | Exp. 2[3] | Exp.1 | Exp. 2 | Exp. 1 | Exp. 2 |
| **Men over age 45 (n=5, Exp. 1; n=4, Exp.2)** | | | | | | |
| 0 | 71±14[4] | 36±18 | 3.7±0.2 | 2.5±0.3 | 33±6 | 20±1 |
| 3 | 60±9 | 34±22 | 3.6±0.6 | 2.6±0.7 | 35±6 | 17±2 |
| P Value | 0.16 | 0.91 | 0.74 | 0.84 | 0.50 | 0.12 |
| **Postmenopausal Women (N=4)** | | | | | | |
| 0 | 78±8 | 47±8 | 3.8±0.2 | 1.8±0.4 | 23±2 | 11±2 |
| 3 | 52±9 | 44±9 | 3.5±0.4 | 1.6±0.5 | 18±2 | 11±3 |
| P Value | 0.02 | 0.37 | 0.58 | 0.19 | 0.26 | 0.86 |
| **Postmenopausal Women on Estrogen Therapy (n=5)** | | | | | | |
| 0 | 61±6 | 33±7 | 2.8±0.5 | 1.7±0.3 | 107±22 | 99±15 |
| 3 | 55±7 | 28±6 | 1.8±0.3 | 1.8±0.2 | 145±36 | 157±27 |
| P Value | 0.02 | 0.24 | 0.08 | 0.27 | 0.09 | 0.02 |
| **Above Combined** | | | | | | |
| 0 | 74±7 | 39±23 | 3.3±0.2 | 1.9±0.8 | 52±12 | 48±13 |
| 3 | 59±5 | 35±26 | 2.8±0.3 | 1.9±1.0 | 64±19 | 69±22 |
| P Value | 0.0008 | 0.31 | 0.06 | 0.92 | 0.13 | 0.06 |

[1]Amount of B as sodium borate supplemented to a diet containing about 0.25 mg/2000 kcal. There was a depletion period of 63 d when no supplement was given followed by a repletion period of 49 d when the supplement was given.
[2]Diet was low in Mg (115 mg/2000 kcal) and marginal in Cu (1.6 mg/2000 kcal); values obtained during the last 42 d of depletion and last 35 d of repletion were compared.
[3]Diet was adequate in Mg (315 mg/2000 kcal) and Cu (2.4 mg/2000 kcal). Values obtained during the last 35 d of each dietary period were compared.
[4]Mean ± SEM.

# B HOMEOSTATIC·MECHANISMS

As stated earlier, one of the criteria often used to support essentiality is homeostatic control mechanisms. Evidence that B is homoeostatically controlled includes the lack of accumulation of B in tissues, the

relatively narrow range of B concentrations in blood of apparently healthy individuals, and the rapid urinary excretion of absorbed B.

Because there is no usable radioisotope of B, the study of its metabolism has been made difficult. However, sodium borate, boric acid, and most likely food B are rapidly absorbed, and excreted mainly in the urine. Most ingested B probably is converted into boric acid, the normal hydrolysis end product of most B compounds and the dominant inorganic species at the pH of the gastrointestinal tract. Thus, B is probably absorbed, transported throughout the body, and excreted mainly as undisassociated boric acid. During transport in the body, boric acid most likely is weakly attached to biosubstances containing *cis*-hydroxyl groups.

In addition to being the major homeostatic mechanism for controlling the body content of B, urinary excretion of B is a good indicator of recent B intake. The mechanism for the movement of B into the urine probably is something more than just the movement of B down a concentration gradient, because the concentration of B in urine can be markedly higher than blood. Hunt and coworkers *(30)* found that in 11 postmenopausal women, increasing dietary B from a deficient 0.36 mg to an adequate 3.23 mg/d increased mean fasting plasma B concentration only from 64 to 95 ng/mL, whereas the mean daily urinary excretion of B increased from 0.37 to 2.87 mg/d without an increase in urine volume. In another study, a B supplement of 2.5 mg/d given to 43 perimenopausal women consuming about 1.2 mg of B/d, based on urinary excretion, increased fasting B concentration from 34 to 53 ng/mL, whereas mean urinary excretion increased from 1.19 to 3.29 mg/d without apparently affecting urine volume *(31)*.

## BIOCHEMICAL FUNCTION OF B

With all the circumstantial evidence for the essentiality of B presented above and by others in this volume, it is surprising that B is not given more consideration as an element of nutritional importance for human health and well-being. As indicated earlier, resistance to this idea probably arises because a defined biochemical function has not been found for B. This lack should not be surprising, because a biochemical function for B in plants has been difficult to define and is only now emerging, almost 75 yr after it was accepted as being essential to complete the life cycle of plants *(32,33)*. A consensus seems to be building for the biochemical role of B being a structural role in cell walls of plants *(34,35)*. However, B is also thought to have a role at the membrane level in plants *(36,37)*. Such a role supports a hypothesis that has been advanced for the biochemical role of B in higher animals *(38)*. This hypothesis is that B has a role in cell membrane function or stability, such that it influences the response to hormone action, transmembrane

signaling, or transmembrane movement of regulatory cations or anions. This hypothesis is supported by the finding that B influences the transport of extracellular Ca and the release of intracellular Ca in rat platelets activated by thrombin *(16)*, and that B influences redox actions involved in cellular membrane transport in plants *(36)*.

Another hypothesis *(18)* that accommodates a large and varied response to B deprivation and the known biochemistry of B is that it acts as a metabolic regulator through forming esters or complexes with a variety of substrate or reactant compounds in which there are hydroxyl groups in favorable positions. Because these complex or ester formations usually result in a competitive inhibition of at least two classes of enzymes in vitro, the regulation by B is hypothesized to be mainly inhibitory.

## DIETARY GUIDANCE FOR B

Because dietary guidance seems justified for B, suggestions for the guidance seems appropriate. In the human depletion–repletion experiments discussed above, the subjects responded to a B supplement after consuming a diet supplying only about 0.25 mg B/2000 kcal for 63 d. Thus, humans apparently have a dietary requirement higher that this. An analysis of both human and animal data resulted in the suggestion that an acceptable safe range of population mean intakes of B for adults could well be 1–13 mg/d *(39)*.

Based on recent analysis of foods and food products, estimations of daily intakes of various age and sex groups have been made. For example, Rainey and coworkers *(40)* reported that the estimated median, mean, and 95th percentile daily intakes of B were 0.79, 0.98, and 2.33 mg/d for adults aged 17 and older. The finding that the median intake was 0.79 mg/d suggests that there may be a significant number of people not consuming B in amounts that could be considered beneficial. This suggestion is supported by the study of a group of 43 perimenopausal women in the eastern North Dakota area of the US *(31)*. Two women apparently consumed an average of <0.5 mg/d of B, and 14 women consumed between 0.5 and 1.0 mg/d of B over a 90-d period based on urinary B excretion data. These findings suggest that B could be a practical nutritional or clinical concern. Based on findings from laboratory studies of animals and humans, B deprivation might be of special concern to people exposed to certain nutritional stressors that are somewhat common today, such as Cu, Mg, and vitamin D deficiencies. Thus, there is a need to increase the awareness of intakes of B that are beneficial to human health and well-being. In other words, there is full justification for sources of food and nutrition information and education (including the appropriate DRI committee) to provide dietary guidance for B.

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