Calcium Bioavailability in Relation to Bone Health

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Abstract: A well established stable isotope technique exists for measuring calcium absorption from single foods and meals, but the long term effects of calcium on bone health cannot be assessed from acute bioavailability studies. Bone health depends primarily on the degree of mineralization, measured as bone mineral density (BMD), and phenotypic variations depend on genetic and environmental factors including calcium supply. Since almost all retained calcium is used for bone mineralization and remodeling, BMD can be used as a long-term (> six months) marker of dietary calcium bioavailability. However, BMD is a very insensitive marker of calcium bioavailability, so its use in dietary intervention studies is restricted to periods of significant bone growth or loss. Biochemical markers of bone metabolism may be used to predict the overall bioavailability of dietary calcium over a shorter time period (> four weeks), but they have a high coefficient of variation, so may not be appropriate for some dietary intervention studies. A group of European laboratories is currently developing an alternative approach using a long-lived radioisotope (^{41}Ca) to label bone calcium and to directly measure the rate of calcium loss from urinary excretion data. The efficiency of calcium absorption is inversely related to intake; whole body balance of the mineral is dependent on rates of absorption and excretion and limited by calcium-binding substances in the gut. Dietary data and indirect measures of bone health indicate that bioavailability is important when habitual intakes are low, especially during periods of bone growth or loss. Further research is required to quantify the effects of major dietary modulators of calcium balance on bone health and to understand their relationship with genetic and physiological variables.

Key words: Calcium, bioavailability, absorption, bone, human nutrition

Introduction

The skeleton contains two types of bone: highly calcified cortical tissue that forms the outer surfaces of most bones and the shafts of long bones, and trabecular tissue that is mainly found at the end of the long bones and in the vertebrae. The growth and maintenance of bone tissue is dependent on a wide range of environmental and genetic factors. The latter accounts for approximately 60–70% of the phenotypic variance in bone mineral density (BMD), whereas environmental factors such as diet and lifestyle account for only 30–40% [1]. There is no evidence, as yet, for a major gene influencing BMD; the familial effects are assumed to be polygenic. Since it is possible to modify environmental variables to improve bone health, it is important to characterize the effect of diet, including the pivotal role of calcium, on bone metabolism.

Osteoporosis is a metabolic bone disease characterized by low bone mass and deterioration of bone tissue that leads to bone fragility and increased risk of fracture. Postmenopausal (Type I) osteoporosis in women aged 50–75 years (incidence ratio of women:men, 6:1) involves loss of trabecular bone and results typically in wrist fracture and spinal vertebrae crush fractures; senile (Type II) osteoporosis in people over 70 years of age (women:men, 2:1) involves loss of both cortical and trabecular bone, and results in hip fracture and spinal vertebrae wedge fractures. Strategies to prevent or delay osteoporosis include maximizing peak bone mass in teenagers and slowing the rate of adult bone loss, especially in peri- and postmenopausal women.

Calcium bioavailability

Calcium balance depends on the relationship between intake (absorption) and excretion (urinary, fecal, and other obligatory losses); a positive balance must be maintained during growth so that calcium accretion in the skeleton can take place. There is wide intersubject variation in calcium absorption, depending on a number of dietary and host-related factors [2]. The latter include age (bone maturity, not chronological age), physiological state (e.g., pregnancy, lactation), estrogen status, intestinal secretions and motility, physical activity (weight-bearing exercise), vitamin D status, and genotype. So far, only one gene polymorphism (vitamin-D-receptor (VDR) gene) has been implicated in modulating the efficiency of calcium absorption [3]. There is no evidence, as yet, for a major gene influencing BMD; the familial effects are assumed to be polygenic. Cortical bone is less influenced by genetic factors than trabecular bone [4], the effect being most important around the age of 26 [5], suggesting that genetic factors have a greater influence on the acquisition of peak bone mass than on bone preservation. Research on the interaction between genotype and diet is in its infancy. There is evidence to indicate that dietary calcium modulates the effect of VDR genotype on bone loss [4, 6], but little is known about the effect of VDR genotype on bone accumulation in children consuming different levels of calcium. A higher self-reported milk consumption in teenage years did not confer a protective effect on fracture risk at ages 40–65 years, but individuals with BB VDR genotype had an increased risk of hip fracture compared with Bb or bb, and the genotype effect was greater in the lower (< 1078 mg/day) calcium group [7]. Further information is required on the effect of genotype on calcium absorption and genotype-dependent responses to calcium supplements.

Calcium is absorbed by active transport and passive diffusion across the intestinal mucosa. Active transport is vitamin D-dependent and accounts for most of the absorbed calcium at low to moderate intakes. Passive diffusion is paracellular and is more important at high intakes. Calcium absorption and intake are inversely related, declining from 45% at intakes of 200 mg/day to 15% at intakes above 2000 mg/day. In women, absorptive efficiency falls with age, declining 2.2% at the time of the menopause and then 0.21% each year thereafter [8]. Efficiency of absorption varies throughout the lifespan, being highest in infancy, rising again in early puberty and mid- to late pregnancy, and declining with age [2]. Changes in calcium intake lead to up- or down-regulation of absorption [9].

The most widely accepted method for assessing the bioavailability of calcium in a single food or meal is a double stable isotope technique derived from a radioisotope method [10]. An intravenous infusion of a calcium stable isotope is given together with the test meal that has been extrinsically labelled with a different calcium stable isotope, and fractional absorption from the oral dose can be calculated from the appearance of the two isotopes in the plasma or urine.

Methods to evaluate bone health

Virtually all calcium retained from the diet is used for bone mineralization and remodeling, thus calcium deposition in bone and its subsequent metabolism could be employed as an indirect long-term assessment of utilization or bioavailability. The measurement of bone mineral status is performed by techniques such as absorptiometry (e.g. DXA), quantitative computerized tomography, and ultrasound, all of which are fairly insensitive and primarily applicable to situations where significant bone deposition or loss is occurring and when modulating factors have a major impact; i.e., the effect is pharmacological rather than nutritional.

Bone resorption and formation are coupled, though separated in time, in a process known as remodeling which ultimately determines bone mass. Clinically, bone biomarkers are used to assess the response to drug therapy by measuring the rates of bone formation and resorption after a minimum of three months of treatment. The percentage change with successful osteoporosis treatment is usually high: 40% to 60% reduction of bone resorption after three to six months [11]. Changes in bone turnover affect calcium metabolism and vice versa. Our understanding of the relationship between nutritional factors and bone health may therefore be advanced by assessing bone biomarkers in response to nutritional interventions. The literature on changes in bone biomarkers in response to nutritional intervention is growing rapidly, however important issues need to be addressed to allow a meaningful assessment of the outcome of these studies. For example what percentage change in biochemical markers demonstrates a positive response to dietary intervention? For clinicians, the recommendation is to consider a reduction of > 30% (80% confidence) as a real change [12]. The large day-to-day variation in urinary markers of bone resorption (> 15%) [13] and wide intersubject variation [14] means that numbers of volunteers needed for studies on biomarker responses to nutrition are relatively high, and dietary intervention studies should be carried out long enough to detect changes in the markers of formation so that a complete assessment of bone turnover may be determined. The other important consideration is the permanency of observed changes in bone remodeling; appropriate study designs must be used to address these issues.

A novel ^{41}Ca radiotracer technique is currently under development for monitoring bone calcium metabolism in response to changes in environmental factors. The isotope has a very long half-life (> 100 000 years) and once introduced into the body, it will label bone calcium for a very long period. The exposure to radioactivity of the small amounts administered is minimal, yet ^{41}Ca released from the skeleton in labeled individuals may be detected in urine by accelerator mass spectrometry (AMS) for many years [15]. Approximately 160 days post-dosing, blood or urine ^{41}Ca tracer reflects the calcium resorption from the labeled part of the skeleton [16]. The longer the individuals are monitored, the more likely it is that ^{41}Ca appearing in urine reflects changes in the whole skeleton rather than the bone sites that are characterized by a high turnover. Once equilibrium has been achieved, it is hypothesized that it will be possible to measure the effects of dietary and other variables on the rate of bone calcium loss, and hence characterize more closely the relationship between diet and bone health. This study technique is still in its infancy and limited to a handful of laboratories because of the few facilities worldwide that can measure ^{41}Ca, and the limited availability of the isotope.

Calcium intake and bone status

Results of observational studies on childhood calcium intake and bone status are inconsistent, probably due to the poor quality of dietary data coupled with lack of quantitative information on calcium bioavailability from different diets [17]. Age, gender, pubertal status, body size, and parental bone values (mothers > fathers) accounted for 80% of the variance in bone mineral content in adolescents [18]; although diet was less important, high calcium intake in girls was associated with a significantly greater increase in bone mineral content and density from 11 to17 years of age. Higher calcium intakes have also been shown to increase exchangeable bone calcium in children aged 5–18 years [19]. Kardinaal *et al* [20] measured radial bone density in girls and young women from six European countries with mean daily calcium intakes ranging from 609 mg in Italy to 1267 mg in Finland. After adjustment for confounders, radial BMD was not found to vary significantly among quartiles of calcium intake for either age group. However, multivariate linear regression demonstrated a weak positive association between calcium intake and BMD in the girls, the association being strongest pre-puberty. In women with a low calcium intake, low fractional calcium absorption has been shown to increase the risk for hip fracture [21].

Results of randomized controlled trials indicate that BMD can be modified by diet. Zamora *et al* [22] reported that vitamin D supplementation in infancy resulted in a higher BMD at specific skeletal sites in pre-pubertal Caucasians. Some studies show that differences in bone accretion rates resulting from calcium supplementation are not persistent and are modest relative to the annual percentage increases in BMD typically observed in pediatric populations. Others demonstrate a persistent effect: for example, 850 mg/day additional calcium from calcium-rich foods increased the femoral shaft BMD of girls [23] and the effect was sustained one year after the intervention. Effects were particularly apparent in individuals with lower self-selected dietary calcium intakes, who also had a greater change in height. Calcium supplementation (1000 mg/day) in female adolescents increased BMD, the effect persisting for at least 14 months [24]; over 12 months, supplementation increased the BMD of 14-year-old girls, the beneficial effect being sustained for at least five years [25].

The influence of childhood calcium intake on adult bone status is more difficult to assess. Retrospective studies report inconsistent findings, mainly due to the difficulties in recalling calcium intakes. The prospective study of Valimaki *et al* [26] suggests that exercise level is the strongest determinant of femoral neck BMD but that women with higher calcium intakes as a child (800–1200 mg/day) have a higher BMD than those with intakes < 800 mg/day. Within the higher calcium intake range (941–1204 mg/day), there is no effect of intake on peak bone mass [27], suggesting that there may be a threshold effect. Indeed, a prospective study investigating adult BMD (DXA) and childhood milk consumption in females with moderate childhood calcium intakes ranging from 986–1157 mg/day, demonstrated a positive relationship between milk intake and adult BMD [28].

Reducing the rate of bone loss is a recognized strategy for improving bone health. Calcium and vitamin D supplementation for one year slowed bone loss from the femoral neck, spine, and whole body in the elderly (> 68 years) but the benefits to bone health had disappeared within two years, emphasizing the need for continuous supplementation [29].

Dietary modulators of calcium bioavailability

Dietary constituents that have been reported to affect calcium absorption and/or excretion are shown in Table I. With respect to putative adverse effects on bone health, only protein, sodium, and caffeine will be discussed as they are probably the most important determinants of calcium balance. Evidence for effects on BMD and fracture risk is shown in Table II.

Table I: Dietary modulators of calcium absorption and excretion

Constituent	Effect	Evidence strong	Evidence moderate	Evidence weak
Protein	Calciuria	II		
Sodium	Calciuria	II		
Caffeine	Calciuria	II		
Phosphorus	Increases endogenous Ca secretion and decreases urinary Ca losses		II	
Fructose	Negative Ca balance			II
Vitamin D	Enhances Ca absorption	II		
Oxalate	Inhibits Ca absorption	II		
Phytate	Inhibits Ca absorption	II		
CPPs*	Enhances Ca absorption			II
NDOs**	Enhances Ca absorption			II

* Caseinophosphopeptides.
** Nondigestible oligosaccharides.

Table II: Dietary modulators of bone health

Constituent	Hypothesis
Animal Protein (and others, e.g. grain products)	Acid precursors; chronic acid stress mobilizes bone and calcium acts as a buffer
Milk	High Ca intake is associated with high BMD
Sodium	Calciuria; increases bone resorption
Caffeine	Calciuria; increases bone resorption
Fruits and Vegetables	Alkaline-producing foods; source of buffer in the diet; reduces urinary calcium loss

a) Protein

Most epidemiological studies indicate that there is an inverse relationship between dietary protein and bone health, as evidenced by the positive association between hip fracture incidence and protein intake; protein stimulates urinary calcium excretion, the mean increase in adults being estimated as 60 mg/day for each 50 g increment in protein intake [30]. An immediate reduction in renal calcium reabsorption has been reported, which may be a response to acid and sulfate generated from sulfur amino acid metabolism. However, the negative effect of protein is probably only significant under conditions of low calcium intake and depends on other constituents [31]. For example, potassium [32] and phosphorus [33] both blunt the hypercalciuric response. This may partly explain the positive effect of fruits and vegetables on bone health. Although the efficiency of absorption of calcium does not increase to offset the losses with high protein intakes, a calcium-to-protein ratio > 20:1 probably provides adequate protection to the skeleton [33].

b) Sodium

The controversy surrounding high dietary salt intakes and the effect on bone health is due to a lack of data on long-term effects on bone metabolism. There is general agreement that short-term high salt intake results in calciuria at low and high calcium intakes in men and women of all ages [34–37]. However, the observation that the calciuric effect might be limited to salt-sensitive individuals [38] warrants further investigation. Recent data published on the effects of salt on bone biomarkers [37, 38] and BMD [39–41] are inconsistent and may be related to differences in study design. Hence, the salt debate in relation to osteoporosis remains controversial.

c) Caffeine

A calciuric effect lasting for several hours has also been demonstrated for caffeine [42], however this has not been tested over several days to allow for adaptation. The analysis of data from a large balance study [43] suggests that an inverse association exists between caffeine intake and calcium absorption efficiency at low and high calcium intake. Although most studies investigating the effects of habitual caffeine intake and fracture risk agree that there is a positive association between the two, the findings from bone mineral density studies are less consistent possibly because of differences in study design and study populations [44].

Calcium supplementation

The bioavailability of a calcium salt is not proportional to its solubility [45] and calcium-citrate-malate has been shown to be superior in bioavailability to other calcium salts [46]. However, given with a meal, calcium carbonate and other calcium salts are as well absorbed as milk calcium [47]. Twice daily administration will enhance efficiency of absorption [48] and calcium supplementation at night suppresses the nocturnal increase in bone resorption markers and reverses the nocturnal increase in parathyroid hormone (PTH) [49]. The effectiveness of various calcium supplements in decreasing bone loss and fracture incidence has been demonstrated in longitudinal studies in postmenopausal women [50, 51].

Conclusions

The relationship between calcium and bone health is very complex, as reflected in the extensive and often conflicting reports in the literature. However, there is convincing evidence that calcium supply affects both the development and rate of bone loss. Higher calcium intakes leading to a more positive calcium balance will increase BMD. Dietary data and indirect measures of bone health indicate that bioavailability is important when habitual intakes are low, especially during periods of bone growth and loss. Further research quantifying the effects of the major dietary modulators of calcium homeostasis on bone health and their dependence on genetic and physiological variables is required.

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