

Evolving story of bone health and the nutritional support

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CALCIUM AND VITAMIN D IN BONE HEALTH

Calcium and vitamin D supplementation is historically one of the most researched and sound nutritional practices to strengthen the bone structure in infants, children, women, men and diverse racial or ethnic groups. Based on the 2010 position statement of The North American Menopause Society adequate calcium and vitamin D intake along with balanced diet, adequate exercise, smoking cessation and avoidance of excessive alcohol intake are the most important measures to maintain bone health and prevent osteoporosis. In one study with human volunteers the inorganic form of calcium, calcium carbonate, and an organic form of calcium, calcium citrate, produced identical 24-hour time increment in total serum calcium levels. Thus, inorganic and organic forms were equally absorbed and had equivalent bioavailability. Given the equivalent bioavailability of the two marketed products, the cost benefit analysis favors the less expensive carbonate product (1).

The effects of vitamin D and calcium cannot be separated. Vitamin D₃ with calcium supplementation compared to placebo has a beneficial effect on bone mass density (BMD) in individuals with osteoporosis, and may reduce the risk of fractures (2). The risk of toxicity with "high" amounts of vitamin D intake is low. To achieve optimal vitamin D status, daily intakes of at least 1000 IU of vitamin D are required (3). Vitamin D₃ is inactive biologically and needs to be changed, or hydroxylated, twice in the body to become active to 1,25-di-hydroxyvitamin D, called calcitriol. Calcitriol, a steroid hormone, is the active form of vitamin D in the body. Calcitriol mediates its biological effects by binding to the vitamin D receptor (VDR), which is located in the intestine, bone,

kidney, and parathyroid gland cells to maintain calcium and phosphorus levels in the blood with the assistance of parathyroid hormone and calcitonin. The binding of calcitriol to the VDR upregulates the gene expression of transport proteins (such as calbindin), which are involved in calcium absorption in the intestine.

A little known finding is that oral bone and tooth loss are correlated with bone loss at non-oral sites. In one study tooth loss was examined in 145 healthy subjects aged 65 years and older who completed a 3-year, randomized, placebo-controlled trial of the effect of calcium and vitamin D supplementation on bone loss from the hip with the 2-year follow-up of the patients (4). During the 2-year follow-up period, 31 of the 77 subjects

ABSTRACT

The research on health of skeletal system while recognizing the important role of calcium and vitamin D₃ goes beyond these nutritional standards in maintaining bone health. Preclinical and clinical studies indicate that essential fatty acids (especially docosahexanoic acid, DHA), carotenoids (such as lycopene), and most significantly vitamin K₂ (menaquinone) may contribute independently and collectively to bone health. Natural Vitamin K₂ as menaquinone MK-7 has been recently clinically demonstrated as having a fundamentally important role in calcium utilization in both bones and the cardiovascular system. Osteocalcin and matrix-GLA protein involved in building bone matrix and keeping calcium from accumulating in the arterial walls respectively need sufficient Vitamin K₂ to function properly. Part of the mechanism of menaquinone MK-7 may be related to preventing excessive expression of the pro-inflammatory factor Nf-kappa-beta and subsequent prevention of osteoporosis due to osteoclast proliferation (osteoclastogenesis).

(40%) with total calcium intake of at least 1000 mg per day lost one or more teeth compared with 40 of the 68 subjects (59%) who consumed less calcium. These findings suggest that intake levels of calcium and vitamin D aimed at preventing osteoporosis have a beneficial effect on tooth retention.

DOCOSAHEXAENOIC ACID (DHA) IN BONE HEALTH

Further studies of the dietary factors affecting bone health show that a higher dietary omega-3 / omega-6 fatty acids ratio is associated with beneficial bone health effects. The omega-3 long-chain variety of fatty acids, especially docosahexaenoic acid or DHA found in fish oil, may play an important role in the regulation of bone metabolism. There are different mechanisms by which dietary fatty acids may improve bone health: their effect on calcium balance, effect on the physiology of bone cells, and concomitant decreases in inflammatory factors that may negatively influence bone build-up and its regeneration such as interleukins and tumor necrosis factor alpha (TNF-alpha) (5).

Preclinical and human studies have indicated that long chain polyunsaturated fatty acids may enhance calcium absorption, reduce urinary calcium excretion, and increase bone calcium content. Significant correlations were found between the docosahexaenoic acid (DHA) supplementation and bone density and bone calcium content in experimental animals (6). DHA supplementation increased absorption of calcium and accrual of calcium in bone significantly more than the eicosapentaenoic acid (EPA) supplementation.

In an epidemiological study, 78 healthy young men with a baseline age of approximately 16 years old were evaluated nutritionally and in bone measurements until the age 24 (7). Bone mineral density of hip and spine were measured at baseline and at 22 and 24 years of age. The subjects' serum levels of omega-3 essential fatty acids were evaluated. A positive correlation was found in course of the study between omega-3 fatty acid serum concentrations and the changes in bone mass density (BMD). Concentrations of DHA were positively associated with total BMD and BMD at the spine. The results indicate that omega-3 fatty acids, especially DHA, are positively associated with bone mineral accrual in young men (7).

Interestingly, supplementation with long-chain essential fatty acids may also play a significant role in prevention of bone loss in periodontal disease, and ultimately prevent the teeth loss. Mice fed

tuna oil and inoculated with *Porphyromonas gingivalis*, a bacterium that causes periodontal disease, showed 72% less alveolar (periodontal) bone loss compared with the control group. The authors of this study concluded that alveolar bone loss may be inversely related to omega-3 polyunsaturated fatty acid tissue levels, and fish oil dietary supplementation may play an important role in periodontal disease prevention and in periodontal disease management (8).

LYCOPENE AND BONE HEALTH

Our understanding of the complex role that nutrition plays in bone health is constantly evolving with increased attention given to new nutrients in addition to the archetypical examples of calcium and vitamin D. Relatively little is known about fruit and vegetable derived carotenoid consumption among postmenopausal women. Carotenoids are tetraterpenoid organic pigments split into two classes, xanthophylls and carotenes which are an important source of antioxidants and vitamin A in the diet; four carotenoids, referred to as provitamin A carotenoids, i.e. beta-carotene, alpha-carotene, gamma-carotene, and beta-cryptoxanthin can be converted in the body to vitamin A. The primary carotenoids in human serum are alpha- and beta-carotene, lycopene, beta-cryptoxanthin, lutein, and zeaxanthin.

One study investigated the relationships between serum carotenoid concentrations, fruit and vegetable intake, and osteoporosis in postmenopausal women (9). It has been found that serum lycopene concentrations were significantly lower in women with osteoporosis as compared to the healthy controls. The total fruit and vegetable intake correlated well with serum levels of lycopene, alpha-carotene, zeaxanthin and beta-cryptoxanthin. Therefore, the carotenoids and specifically lycopene that apparently is lower in women with osteoporosis as compared to healthy controls may play a beneficial role in maintaining healthy bone structure and preventing osteoporosis.

In a randomized, placebo controlled intervention study, sixty postmenopausal women supplemented with lycopene were evaluated nutritionally and in the oxidative stress parameters (10). The patients were divided into groups that received i) regular tomato juice, ii) lycopene-rich tomato juice, iii) tomato Lyc-O-Mato® lycopene capsules, or iv) placebo capsules, twice daily for total lycopene intakes of 30, 70, 30, and 0 mg/day respectively for 4 months. The lycopene supplementation, regardless of the source of supplement, for 4 months

significantly increased serum levels of this carotenoid, as compared to the placebo receiving group, and also resulted in significantly increased antioxidant capacity and decreased lipid peroxidation, protein oxidation, and markers of bone resorption. These laboratory findings suggest that postmenopausal women supplemented with lycopene may benefit significantly with increased antioxidant capacity and related decrease in bone resorption, as assessed by decrease in levels of bone resorption marker.

Another intervention study has examined the influence of individual carotenoid intake on the risk of bone fracture in elderly men and women (11). The intake of alpha-carotene, beta-carotene, beta-cryptoxanthin, lycopene, lutein and zeaxanthin on the frequency of hip fracture and non-vertebral osteoporotic fracture was evaluated in 370 men and 576 women with a mean age of 75. The study population completed a food frequency questionnaire in 1988-1989 and were monitored for hip fracture until 2005 and nonvertebral fracture until 2003. A total of 100 hip fractures occurred over 17 years of follow-up. Subjects with the highest total carotenoid intake had significantly lower risk of hip fracture, and subjects with higher lycopene intake had significantly lower risk of hip fracture and non-vertebral fracture. A non-significant protective trend was observed for total beta-carotene intake for hip fracture alone. No significant associations were observed between the incidence of fractures and levels of intake of alpha-carotene, beta-cryptoxanthin, or lutein and zeaxanthin. These results suggest a protective role of lycopene and several other carotenoids for bone health in older adults.

The relationships of dietary intakes of retinol and carotenoids were examined in an Anglo-Celtic Australian population of 68 men and 137 women (12). Bone mass of total body and lumbar spine were positively related to lycopene intake in men, and to lycopene and lutein/zeaxanthin intake in premenopausal women. In addition, a positive association of lumbar spine bone mass with dietary beta-carotene intake was observed in postmenopausal women. No relationship was found between dietary retinol intake and bone mineral status. The finding of this study suggests that fruit, vegetable consumption and dietary carotenoid intake has a beneficial effect on bone health.

VITAMIN K2

The nutritional role of menaquinones or vitamin K2 is increasingly recognized and

distinguished from the biological role of vitamin K1 or phyloquinone (13). Epidemiological studies show that dietary intake of natto, a traditional food in Japan prepared from fermented soy-beans which contains significant amount of menaquinone-7 (MK-7), reduces the risk of bone mass loss and/or age related decline in bone tensile strength. Vitamin K2, especially MK-7 form, with the half life in plasma significantly longer compared to phyloquinone, plays an important role as a co-substrate for the enzyme gamma-glutamyl carboxylase which carboxylates glutamic moiety of certain biologically important proteins (13). Vitamin K2 is responsible for carboxylation and activation of osteocalcin, which is specific to bone gamma-carboxy-glutamic acid protein synthesized by bone building cells osteoblasts.

The bone is constantly remodeled and the remodeling is mediated by two cell types, osteoblasts (bone building) and osteoclasts (bone resorption). Osteoblastic bone formation and osteoclastic bone resorption are reflected in levels of bone metabolism markers, e.g. intact osteocalcin, carboxylated osteocalcin, under-carboxylated osteocalcin and bone alkaline phosphatase, an enzyme involved in bone mineralization. Bone resorption can also be evaluated by measuring the urinary free and total pyridinoline, a molecule that cross-links collagen in the bone structure. The cross-links are degraded by the lysosomal enzymes of osteoclasts during bone resorption which results in release of deoxypyridinoline, a marker of bone resorption detected in urine.

In one clinical study the serum level of undercarboxylated osteocalcin – indicating biologically inadequate osteocalcin – and levels of bone turnover markers, i.e. intact osteocalcin (OC), bone alkaline phosphatase (BAP) and urinary deoxypyridinoline (DPD) were evaluated in early postmenopausal women receiving Vitamin K2 and Vitamin D3 for up to 24 months (14). Thirty-four

postmenopausal women with a mean age of 53 with osteoporosis and low bone mineral density were enrolled into the study. The serum levels of under-carboxylated osteocalcin in women treated with vitamin K2 alone and with vitamin K2 and vitamin D3 decreased significantly. Serum levels of intact OC and BAP in women treated with vitamin K2 did not show significant changes, while the levels in women who received the combined treatment decreased significantly. The urinary DPD level in women treated with vitamin K2 did not change, while in women who received the combined treatment the DPD marker tended to decrease. Therefore combined treatment with vitamin K2 and vitamin D3 may be more effective for sustaining bone health in early postmenopausal women than vitamin K2 stand alone supplementation.

The recent epidemiological findings indicate that there may be an additional rationale in supplementing vitamin K2 to postmenopausal women to prevent osteoporosis. In an article published in *British Medical Journal* the 1,471 healthy postmenopausal women were divided into two study groups, one group given 1 g daily of calcium supplements and the other placebo (15). The study concluded that patients taking calcium were more than twice as likely to have heart attacks as patients on placebo. This finding may show that increased calcium use to strengthen bones – but without the necessary co-factors like vitamin K2 – may actually lead to increased risk of cardiovascular events. This phenomena can be better understood since K2 helps in carboxylation and activation of another protein besides osteocalcin, matrix GLA-protein (MGP), for elasticity and prevention of calcification of blood vessels. This dual function of vitamin K2 is sometimes referred to as the vitamin K paradox: vitamin K2 helps build calcium deposits in the bone via carboxylated osteocalcin, while it has the opposite effect on the circulatory system by activating the MGP which prevents

calcium deposits in the arteries and is one of the most potent arterial calcification inhibitors known. It has been suggested that many populations are sub-clinically deficient in vitamin K2 and this deficiency combined with trending calcium supplements may predispose to cardiovascular disease. In addition, epidemiological studies in Netherlands suggest an association between poor vitamin K2 (menaquinone) but not vitamin K1 (phyloquinone) status and increased risk of coronary heart disease, aortic calcification and all-cause mortality (16).

Further understanding of vitamin K2 function comes from its mechanism in bone resorption, especially as it occurs in osteoporosis (17). The menaquinone-7 (MK-7), has been shown to potentially prevent bone resorption by up-regulating osteoprotegerin a decoy receptor for a ligand, which otherwise would attach to receptor activator of NF-κB (RANK) and activate nuclear factor kappa beta (NF-κB). Activation of NF-κB, often referred to as “master switch” of inflammation, is a necessary step for proliferation of osteoclasts and osteoporosis. Therefore by occupying the receptor activator of NF-κB (RANK), vitamin K2 sequesters RANK receptor and prevents further steps leading to formation of osteoclasts or osteoclastogenesis and excessive bone destruction – osteoporosis.

In principle the described action of K2 is anti-inflammatory and similar to the mechanism of natural anti-inflammatory compounds boswellic acids, i.e. acetyl-keto beta-boswellic acid (AKBA) prevents receptor activator of NF-κB ligand (RANKL) signaling, NF-κB activation and osteoclastogenesis (18) (Figure 1).

A new paradigm emerges in nutritional support of healthy skeletal system which i) makes bone health an integral part of a general health and well-being; ii) advocates use of multiple synergistic nutrients; iii) takes into account a complex interaction between nutrients, e.g. nutrients affecting bone and cardiovascular health; iv) considers role of nutrients modifying the inflammatory processes in bone physiology and pathology; and v) recognizes that deficiencies in nutrients other than calcium maybe responsible for poor bone structure and performance.

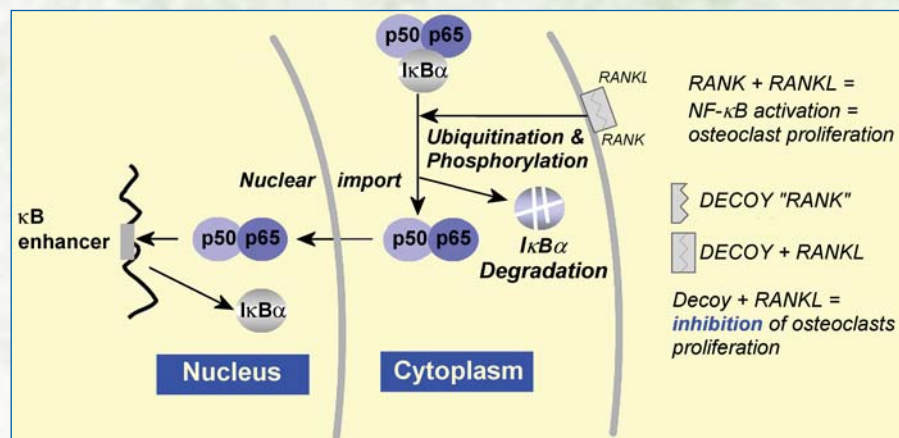


Figure 1 – NF-κB role in osteoclastogenesis

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