Vitamin K in the treatment and prevention of osteoporosis and arterial calcification

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Purpose. The role of vitamin K in the prevention and treatment of osteoporosis and arterial calcification is examined.

Summary. Vitamin K is essential for the activation of vitamin K-dependent proteins, which are involved not only in blood coagulation but in bone metabolism and the inhibition of arterial calcification. In humans, vitamin K is primarily a cofactor in the enzymatic reaction that converts glutamate residues into γ-carboxyglutamate residues in vitamin K-dependent proteins. Numerous studies have demonstrated the importance of vitamin K in bone health. The results of recent studies have suggested that concurrent use of menaquinone and vitamin D may substantially reduce bone loss. Menaquinone was also found to have a synergistic effect when administered with hormone therapy. Several epidemiologic and intervention studies have found that vitamin K deficiency causes reductions in bone mineral density and increases the risk of fractures. Arterial calcification is an active, cell-controlled process that shares many similarities with bone metabolism. Concurrent arterial calcification and osteoporosis have been called the "calcification paradox" and occur frequently in postmenopausal women. The results of two dose–response studies have indicated that the amount of vitamin K needed for optimal γ-carboxylation of osteocalcin is significantly higher than what is provided through diet alone and that current dosage recommendations should be increased to optimize bone mineralization. Few adverse effects have been reported from oral vitamin K.

Conclusion. Phytonadione and menaquinone may be effective for the prevention and treatment of osteoporosis and arterial calcification.

Index terms: Calcinosis; Combined therapy; Dosage; Drug interactions; Estrogens; Mechanism of action; Menaquinone; Osteoporosis; Phytonadione; Toxicity; Vitamin D; Vitamins

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Osteoporosis and arterial calcification are major health concerns in modern societies. It is estimated that 30% of postmenopausal Caucasian women in the United States have osteoporosis and 54% have osteopenia[1,2] and 75–95% of men and women have some degree of coronary artery calcification on autopsy.[3,4] Although osteoporosis and arterial calcification were once thought to be unrelated conditions, recent studies suggest there may be a connection.[5-8] It appears that a common factor in the development of these two disorders may be vitamin K deficiency.

Over the past 20 years, several vitamin K-dependent (VKD) proteins have been discovered. Recent studies have shown that, in addition to their role in carboxylating coagulation factors, VKD proteins are involved in bone metabolism and the inhibition of arterial calcification. The two VKD proteins examined in this review are osteocalcin and matrix Gla protein (MGP). Osteocalcin appears to play a key role in bone metabolism but its mechanism of action has not been fully elucidated.[1] Osteocalcin is synthesized mainly by osteoblasts and, when carboxylated, has molecular properties that allow it to tightly bind hydroxyapatite in bone, thereby promoting mineralization.[9-11] Vitamin K is thought to promote bone mineralization by enhancing the carboxylation of osteocalcin.[10,12,13]

MGP is synthesized primarily by chondrocytes and vascular smooth muscle cells.[14-17] Recent animal studies have shown that MGP plays a key role in the inhibition of tissue calcification.[14,18] As with all VKD proteins, MGP must be carboxylated to function properly.[16,19]

Vitamin K

Vitamin K refers to a group of re-

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lated compounds. There are two natural forms: phylloquinone (vitamin K_1) and menaquinone (vitamin K_2). Phylloquinone is the most common form of vitamin K and is found in leafy green vegetables (e.g., lettuce, broccoli, spinach, cabbage) and vegetable oils (e.g., soybean and canola oils). Commercially prepared vitamin K_1 (phytonadione) is chemically identical to naturally occurring vitamin K_1 (phylloquinone).

Menaquinone includes a range of related forms generally designated as menaquinone-*n* (MK-*n*), where *n* is the number of isoprenyl groups. Menaquinone is found in meat, fermented products, and cheese. The menaquinones most commonly found in food are MK-4, which is a short-chain menaquinone, and the long-chain menaquinones MK-7, MK-8, and MK-9.[20] Intestinal bacteria also produce the longer-chain menaquinones (MK-7–MK-10)[21]; however, bacteria-derived menaquinone appears to contribute minimally to overall vitamin K status.[1,22,23] Menaquinone, in the form of MK-4 (also known as menatetrenone), has been used in Japan for the treatment of osteoporosis since 1995. Dietary supplements containing up to 15 mg of menaquinone (MK-4) per capsule have recently become available in the United States.

In humans, vitamin K is primarily a cofactor in the enzymatic reaction that converts glutamate residues into γ-carboxyglutamate residues in VKD proteins.[21,24-28] Vitamin K deficiency can lead to suboptimal γ-carboxylation of these proteins and impairment of their function.[29-32] These VKD proteins are involved in such functions as coagulation-factor activation (factors V, VII, and X; prothrombin; and fibrinogen), bone metabolism, and inhibition of vascular calcification. The vitamin K requirement for carboxylation of bone and arterial wall VKD proteins is higher than that for the carboxylation of coagulation factors in the liver.[33] Daily vitamin K requirements for maximal γ-carboxylation of the extrahepatic VKD proteins may be significantly higher than recommended by current dietary guidelines.[33] Vitamin K deficiency, resulting in the undercarboxylation of specific VKD proteins, may be an independent risk factor for osteoporosis and arterial calcification.

Vitamin K for osteoporosis: Rationale

A number of human-cell studies have helped define the role of vitamin K and osteocalcin in bone health. Both phylloquinone and menaquinone promote bone mineralization; however, menaquinone has been shown to be more potent,[34] likely because of the enhanced carboxylation of osteocalcin by menaquinone. Menaquinone has been shown to increase osteocalcin accumulation on the cell layer.[34] In vitro, phylloquinone and menaquinone (MK-4) inhibit osteoclast formation and induce the differentiation of osteoprogenerator cells into osteoblasts.[35]

The results of recent human and animal studies have suggested that concurrent use of menaquinone and vitamin D may substantially reduce bone loss.[9,36,37] In rats with ovariectomy-induced bone loss, menaquinone and vitamin D_3 had a synergistic effect on bone-loss reduction.[9] Hirano and Ishii[37] found that the coadministration of calcium, menaquinone, and vitamin D in rats increased the peak bone mass and reduced the loss of bone mineral density (BMD).

Vitamin K for osteoporosis: Clinical studies

The relationship between dietary vitamin K intake and bone status has been investigated in several epidemiologic (Table 1) and intervention studies (Table 2). These studies suggest that vitamin K deficiency causes reductions in BMD and increases the risk of fractures, resulting from the undercarboxylation of osteocalcin. Low intakes of vitamin K have been associated with an increased risk of hip fractures. In a study of 72,327

Table 1.

Epidemiologic Studies of Vitamin K and Bone Health[a]

Ref.	Subjects	Variables Studied	Outcome
8	113 postmenopausal women	UcOC, BMD	UcOC and BMD inversely related
38	72,327 women	Vitamin K intake, hip fracture rate, BMD	Vitamin K intake of >109 μg/day reduced risk of hip fracture by 30%; no correlation between vitamin K and BMD
39	888 men and women	Vitamin K intake, hip fracture rate, BMD	65% reduced risk of fractures in highest quartile of vitamin K intake compared with lowest quartile
40	104 elderly women with hip fractures; 255 controls	UcOC, hip fracture rate, BMD	UcOC (not total OC) predicted fracture risk independently of femoral BMD
41	195 elderly women[b]	UcOC, hip fracture rate	Fracture risk 5.9 times higher in women with elevated UcOC at start of study
42	183 elderly women[c]	UcOC, hip fracture rate	Fracture risk 3.1 times higher in women with elevated UcOC at start of study
43	212 women	UcOC, BMD	UcOC was independent marker for BMD in women 1–10 yr postmenopause

[a] UcOC = undercarboxylated osteocalcin level, BMD = bone mineral density, OC = osteocalcin level.

[b] 18-month prospective study.

[c] Three-year follow-up study.

Table 2.
Intervention Studies of Vitamin K's Effect on Bone Variables[a]

Ref.	Subjects	Treatment	Outcome
12	92 postmenopausal women with osteoporosis	Group D: Vitamin D_3 0.75 μg/day Group K: Menaquinone 45 mg/day Group DK: D and K treatment Group C: Calcium 2 g/day All treatments for 2 yr	BMD improved more in group DK than in group D or group K; increase in lumbar spine BMD significantly greater in group DK than group C
29	219 healthy men and women	Phytonadione 1 mg/day for 2 wk	In all treated groups, mean UcOC decreased from 7.6% to 3.4%; age and sex did not affect the decrease
32	100 healthy adults	Phytonadione 250, 375, 500, or 1000 μg/day or placebo for 2 wk	Phytonadione 1000 μg/day produced greatest carboxylation of OC
33	21 healthy older women	Phytonadione 18 μg/day for 4 wk, then 86, 200, and 450 μg/day, each for 2 wk	Carboxylation of OC not restored by phytonadione 450 μg/day
44	23 postmenopausal women	Phytonadione 80 μg/day, vitamin D_3 350–400 units/day, both, or placebo for 1 yr	Phytonadione at study dosage needed to attain premenopausal %carbOC
46	46 women with osteoporosis	Menaquinone 45 mg/day for 2 yr	Treated group had fewer new vertebral fractures (13) than placebo group (30)
47	20 elderly women with osteoporosis	Calcium 200 mg/day for 2 wk with or without menaquinone 45 mg/day	Menaquinone group had reduction in UcOC without change in OC
48	20 postmenopausal women	Phytonadione 1 mg/day for 2 wk with or without vitamin D_2 400 units/day	Carboxylation of OC improved in both groups; vitamin D_2 had no effect
49	113 women with fractures and 91 women without fractures	Menaquinone 45 mg/day, vitamin D_3 1 μg/day, or both for 4 wk	UcOC decreased in menaquinone groups but not group receiving vitamin D_3 only
54	94 postmenopausal women with osteoporosis (84 controls, 10 treated)	Menaquinone 45 mg/day with either conjugated estrogens 0.625 mg/day or medroxyprogesterone acetate 2.5 mg/day for 1 yr	Menaquinone–hormonal treatment improved BMD that had been decreasing during hormonal treatment alone

[a] BMD = bone mineral density, UcOC = undercarboxylated osteocalcin level, OC = osteocalcin level, %carbOC = percentage of total osteocalcin that is carboxylated.

women, vitamin K intakes (assessed through the use of a food-frequency questionnaire) were inversely related to the risk of hip fracture.[38] The adjusted relative risk (RR = 0.70; 95% confidence interval [CI], 0.53–0.93) of hip fracture was 30% less in the women from the top four quintiles of vitamin K intake (>109 μg daily) compared with women from the lowest quintile (<109 μg daily). This finding is supported by a study of 888 men and women from the Framingham Heart Study.[39] Patients with the highest quartile of vitamin K intake (median, 254 μg daily) had a 65% lower adjusted RR (RR = 0.35; 95% CI, 0.13–0.94) of hip fracture than did those in the lowest quartile of intake (median, 56 μg daily).

Undercarboxylated osteocalcin and bone health. Numerous studies have shown that an association exists among undercarboxylated serum osteocalcin, BMD, and fracture rate.[40-44] In a study of 359 independently-living women, increased levels of undercarboxylated osteocalcin were associated with increased risk of hip fracture, with an odds ratio of 1.9 (95% CI, 1.2–3.0).[40] In a series of reports involving institutionalized elderly women, a strong correlation was found between undercarboxylated serum osteocalcin levels and the subsequent risk of hip fracture.[41] Women with abnormally high undercarboxylated osteocalcin concentrations (>1.65 ng/mL) had a RR between 3.1 (99.9% CI, 1.7–6.0; $p < 0.001$)[42] and 5.9 (99.9% CI, 1.5–22.7; $p < 0.001$) times higher than those with normal undercarboxylated osteocalcin levels (<1.65 ng/mL). Knapen et al.[43] conducted a cross-sectional study of 212 women and found a strong inverse correlation (adjusted RR = 0.5–0.7) between serum undercarboxylated osteocalcin levels and BMD in postmenopausal women. In a trial of 141 postmenopausal women, the percentage of carboxylated osteocalcin to total osteocalcin was measured.[44] The value of that variable was positively correlated with BMD of the lumbar spine ($r = 0.32$, $p < 0.005$) and femoral neck ($r = 0.25$, $p < 0.005$).

Hodges et al.[45] demonstrated that depressed serum levels of phylloquinone and menaquinone (for the latter, most notably MK-7 and MK-8) are found in patients with osteoporotic fractures and suggested that serum levels of phylloquinone and menaquinone can serve as markers for osteoporotic fracture risk.

Vitamin K, osteocalcin carboxylation, and bone health. A number of clinical studies have been conducted investigating the effect of vitamin K administration on the carboxylation of osteocalcin, BMD, and fracture rates.[29,36,46,47] Various dosages of both phylloquinone and menaquinone have been used in clinical trials; however, in all studies, undercarboxylated osteocalcin levels declined significantly with vitamin K supplementation. In a study to determine the prevalence of suboptimal carboxylation of osteocalcin in healthy North American adults, Binkley et al.[29] conducted a single-blind, placebo-controlled trial with 219 healthy young and elderly adults. The treatment group received 1 mg of phytonadione daily for two weeks. At the end of the study, patients receiving phytonadione had a significant decrease in the mean percentage of undercarboxylated osteocalcin, from 7.6% to 3.4% ($p < 0.001$), without significant differences when stratified by age or sex. In a randomized, open-label, controlled trial of 241 Japanese postmenopausal osteoporotic women, the treatment group received 45 mg of menaquinone daily for two years.[46] At the end of the study, the undercarboxylated serum osteocalcin concentrations in the treatment group were significantly lower than in the control group (1.6 ± 0.1 ng/mL and 3.0 ± ng/mL, respectively) ($p < 0.0001$). In addition, the occurrence of fracture in the treatment group was significantly lower than in the control group ($\chi^2 = 10.935$, $p = 0.0273$). In a smaller, randomized, open-label study of Japanese osteoporotic women, Miki et al.[47] found that undercarboxylated serum osteocalcin levels could be reduced in as little as two weeks. The treatment group received 45 mg of menaquinone (specifically MK-4) plus 200 mg of calcium daily. The control group received only 200 mg of calcium daily. After two weeks, the mean ± S.D. serum undercarboxylated osteocalcin concentrations in the treatment group declined from a baseline value of 2.8 ± 0.9 ng/mL to 1.7 ± 0.5 ng/mL ($p < 0.05$). No significant changes occurred in the control group over the same period.

Involvement of vitamin D. It appears that adequate levels of both vitamins D and K may have additive effects in improving bone health. Many studies have investigated the combined effects of vitamins D and K.[12,36,44,48,49] An excellent review on the additive effects of vitamin D_3 and menaquinone was recently published by Iwamoto and colleagues.[50] 1,25 $(OH)_2$ D_3 is the most active vitamin D_3 metabolite and promotes intestinal absorption of calcium, reduces serum levels of parathyroid hormone, and activates the synthesis of osteocalcin.[49,51-53]

In a three-year, randomized, double-blind trial of 155 postmenopausal women, Braam et al.[36] found that vitamin D and phytonadione had a complementary effect on the attenuation of bone loss. Participants were divided into three groups: (1) placebo, (2) vitamin D and mineral (8 μg of vitamin D, 500 mg of milk-derived calcium, 150 mg of magnesium, and 10 mg of zinc daily), and (3) vitamin D, mineral, and phytonadione (same as vitamin D and mineral group plus 1 mg of phytonadione daily). After three years, patients who received vitamin D, minerals, and phytonadione demonstrated a 1.7% reduction (95% CI, 0.35–3.44%) in bone loss from the femoral neck (absolute bone loss of 3.3%) compared with the placebo group (absolute bone loss of 5.0%) and a 1.3% reduction (95% CI, 0.10–3.41%) compared with those receiving only vitamin D and mineral supplements (absolute bone loss of 4.6%). No significant differences were observed among the three groups with respect to changes in BMD of the lumbar spine.

In a two-week, single-blind study, 20 postmenopausal, osteoporotic women were given either 1 mg of phytonadione daily or 1 mg of phytonadione plus 400 IU of vitamin D_2 daily.[48] The mean carboxylation level of osteocalcin was corrected to premenopausal levels (~72%) in both groups, but the addition of vitamin D_2 had no effect on study results. The percentage of carboxylated osteocalcin increased from 57% before treatment to 73% after treatment ($p < 0.001$). A similar finding was reported in a study conducted by Takahashi et al.[49] In that open-label trial of 113 osteoporotic women with femoral hip or vertebral fractures and 91 premenopausal and postmenopausal women without fractures or osteoporosis, participants were randomized to receive menaquinone (45 mg daily), vitamin D_3 (1 μg daily), or menaquinone (45 mg daily) plus vitamin D_3 (1 μg daily) for four weeks. Significant decreases occurred in undercarboxylated serum osteocalcin levels in the menaquinone only ($p = 0.0001$) and the menaquinone plus vitamin D_3 ($p = 0.0018$) groups but not in women treated with vitamin D_3 only.

In a randomized, double-blind study investigating the effects of vitamin D_3 and phytonadione in postmenopausal women, Schaafsma et al.[44] found that a daily intake of 80 μg of phytonadione was necessary to reach premenopausal percentages of carboxylated osteocalcin. At the end of the study, improvements in the percentage of carboxylated osteocalcin were seen in both the phytonadione-treated group with normal BMD ($p = 0.001$) and the phytonadione-treated group with low BMD ($p \leq 0.0001$), compared with the control group, who received no phytonadione. Surprisingly, the percentage of carboxylated osteocalcin also increased in those receiving vitamin D_3 only ($p \leq 0.006$). Another randomized, open-label study supporting the combined effects of vitamin D_3 and menaquinone on BMD in osteoporotic women was conducted by Iwamoto et al.[12] Ninety-two postmenopausal women with osteoporosis were given vitamin D_3 (0.75 μg), menaquinone (45 mg daily), vitamins D_3 (0.75 μg daily) plus menaquinone (45 mg daily), or calcium (2 g daily). After two years, BMD increased significantly in the vitamin D_3- and menaquinone-treated groups, compared with the calcium-treated group ($p < 0.05$ and $p < 0.001$, respectively). However, the most significant increase in BMD was seen in the vitamin D_3 plus menaquinone group ($p < 0.0001$).

Menaquinone was also found to have a synergistic effect when administered with postmenopausal hormone therapy.[54] Hormone therapy is known to increase BMD for two to three years after menopause and maintain it thereafter. For some women taking hormone therapy, the increase in BMD reaches a plateau and then declines. A combined administration of menaquinone (45 mg of MK-4 daily) and hormone therapy was investigated in 10 women who had declining BMD levels. The mean ± S.D. rate of change in their BMD increased significantly, from –2.4% ± 2.5% to 6.7% ± 2.9% ($p < 0.03$) after 12 months of combination therapy.

Two recent studies have provided dose–response data on phytonadione that indicate that current dietary intake recommendations may be inadequate. Binkley et al.[32] conducted a single-blind, placebo-controlled trial to identify the lowest dosage of phytonadione needed to maximally carboxylate osteocalcin. One-hundred healthy adults age 19–36 years were randomly assigned to receive placebo or 250, 375, 500, or 1000 μg of phytonadione daily for two weeks. The percentage of undercarboxylated serum osteocalcin decreased with increasing dosages ($p < 0.0001$), with the greatest reduction occurring in those who received 1000 μg daily. In an 84-day depletion and repletion study, 21 older women received a phylloquinone-restricted diet (18 μg daily), followed by a stepwise repletion of 86, 200, and 450 μg of phytonadione.[33] Various markers of vitamin K status were evaluated to measure participants' response. The carboxylation of prothrombin was restored to prestudy levels with an intake of 200 μg daily. However, carboxylated osteocalcin remained below normal levels after supplementation of up to 450 μg of phytonadione daily. The efficacy of phytonadione and menaquinone supplementation in the treatment of osteoporosis is currently under study in the United States.[55]

Arterial calcification

Similarity to bone metabolism. Arterial calcification was once thought to be a passive process that occurred in response to tissue injury. Evidence now suggests that it is an active, cell-controlled process that shares many similarities with bone metabolism.[3,21,56,57] Arterial calcification occurs at two sites in the vessel wall: the media and the intima. Medial calcification is known as Mönckeberg's arteriosclerosis. Unlike intimal calcification, Mönckeberg's arteriosclerosis occurs independently of atherosclerosis.[21,58,59] Although the cells involved differ depending on the site of calcification, MGP is expressed at both sites.[3] The connection between MGP and vascular calcification has been documented by both cell culture and animal studies. Human cell studies have shown that normal vascular smooth muscle cells express MGP and, at sites of calcification, it is substantially upregulated.[56,58,60] This may be a physiological attempt to minimize calcification and tissue damage.[21,61] In an animal cell study, it was found that endochondral calcification triggered by warfarin could be inhibited by an overexpression of MGP.[15] Animal studies of MGP-deficient mice and warfarin-treated rats provide strong evidence that MGP is a potent inhibitor of vascular calcification.[14,18] Extensive arterial calcification was found in animals that do not express MGP and those that could not carboxylate it. Assuming that the VKD γ-carboxylation of MGP is essential for its inhibitory effect on calcification, vitamin K deficiency may be a risk factor for vascular calcification.

Clinical studies. Concurrent arterial calcification and osteoporosis have been called the "calcification paradox"[3] and occur frequently in postmenopausal women.[6] However, only a few human studies investigating the connection between these two diseases have been published. In a study of 45 postmenopausal women with normal and low BMD, an inverse relationship was found between BMD and coronary calcification.[5] The mean ± S.D. total coronary calcium score (a measure of calcification) was significantly higher in all arteries of the women with low BMD than in the control group (221.7 ± 355.4 and 41.9 ± 83.1, respectively) ($p < 0.025$). In a nine-year, population-based study of 236 women, progression of atherosclerotic calcification was associated with increasing bone loss.[6] Women with progressive aortic calcification had an average loss of metacarpal bone density of 7.2 mm^2%, compared with 5.6 mm^2% in women without progressive aortic calcification ($p < 0.05$). In two studies by Jie et al.,[7,8] a correlation was found between vitamin K status and mineralization of both bone and vessel walls. In a population-based study, vitamin K status, osteocalcin levels, and aortic calcifications were assessed in 113 postmenopausal women.[8] The presence of aortic calcification was associated with lower bone mass and a marginal vitamin K status. There was a 7% difference in the bone mass between women with and without aortic calcification (mean difference, 3.2 mm^2; 95% CI, –0.2 to 6.5 mm^2; $p = 0.06$). In another cohort study, 113 postmenopausal women with aortic calcifications were found to consume approximately 42.9 μg less dietary vitamin K than participants without aortic calcification (95% CI, –6.6 to 92.5 μg).[7] These women also had a 0.32-ng/mL higher adjusted undercarboxylated osteocalcin level (95% CI, 0.03–0.61 ng/mL) and lower hydroxyapetite-binding capacity than those women without aortic calcification.

In a population study of 4500 elderly patients, an inverse relationship was demonstrated between dietary intake of menaquinone and aortic calcification, myocardial infarction, and sudden cardiovascular death.[62]

The most convincing data to date were recently reported by Braam et al.[63] In this three-year, randomized, placebo-controlled trial of 181 healthy, Caucasian postmenopausal women, daily supplementation with 1 mg of phytonadione (plus 8 μg of vitamin D_3 and minerals [500 mg of calcium, 150 mg of magnesium, and 10 mg of zinc per day]) inhibited the loss of carotid artery elasticity, compared with those receiving placebo and those in the vitamin D_3 plus minerals group (–13.2 kPa; 95% CI, –35.8 to –5.3) ($p < 0.01$). No significant differences in arterial elasticity were noted between the placebo group and the vitamin D_3 plus minerals group. In this study, no significant differences were observed in carotid artery intima–media thickness between the three study groups.

Interestingly, no prospective interventional studies have been published investigating menaquinone deficiency as a risk factor for arterial calcification or whether supplementation with menaquinone affects arterial elasticity, arterial calcification, or intima–media thickness.

Adverse effects and dosage of vitamin K

Few adverse effects have been reported from oral vitamin K.[64-66] In the articles reviewed, there was no evidence of toxicity associated with the intake of phytonadione or menaquinone even at a daily dose of the latter of 45 mg.

Adequate intake of phylloquinone for the carboxylation of blood coagulation factors was recently set at 90 μg per day for adult women and 120 μg daily for adult men, based on median dietary intake data from the Third National Health and Nutrition Examination Survey.

In the treatment of osteoporosis, Schurgers and Vermeer[20] suggested a phytonadione dosage of 1000 μg daily. Braam et al.[63] used the same daily dose to inhibit the loss of carotid artery elasticity. However, several trials of osteoporotic, postmenopausal women have used menaquinone dosages as high as 45 mg per day.[12,46,47,49]

Because of the very low toxicity of phytonadione and menaquinone, a 1000-μg daily dose of each is warranted. Doses of menaquinone exceeding 1 mg are not readily available in the United States at this time, but it is being produced in 15-mg capsules by a Canadian nutraceutical company.

Discussion

Over the past decade it has become evident that vitamin K plays a far greater role in human health than previously thought. Vitamin K is essential for the activation, via γ-carboxylation, of VKD proteins. These proteins have various functions and are found throughout the body. Some of these proteins, such as those involved in blood coagulation, have been thoroughly researched. Others, notably those involved in bone metabolism and the inhibition of arterial calcification, have drawn new attention to vitamin K.

Numerous studies have demonstrated the importance of vitamin K in bone health. Cell studies have helped delineate the mechanism by which menaquinone promotes bone mineralization and inhibits resorption.[35] Human and animal studies have clearly demonstrated that vitamin K can improve bone health by increasing bone mass and reducing bone loss.[12,37,46,54]

The results of two dose–response studies have indicated that (1) the amount of vitamin K needed for optimal γ-carboxylation of osteocalcin is significantly higher than what is provided by diet alone and (2) there is a need to increase current dosage recommendations to optimize bone mineralization.[32,33]

The combination of menaquinone and vitamin D_3 has additive beneficial effects on sustaining lumbar BMD and preventing osteoporotic vertebral fractures in postmenopausal women with osteoporosis.[12,50]

The role of vitamin K in the prevention of arterial calcification is not as well researched. Several epidemiologic studies,[5-8] as well as a recent clinical trial of postmenopausal women,[63] have implicated phylloquinone deficiency as a risk factor for arterial calcification and have alluded to a connection between the deficiency and osteoporosis.

Additional research is needed to address several important questions: What is the optimal intake of phytonadione, menaquinone, and vitamin D_3 to support bone mineralization and reduce fracture risk? What is the optimal dosage of phytonadione and menaquinone to preserve the elasticity of arterial endothelial tissue by decreasing the calcification of the intima and media? Do phytonadione, menaquinone, and vitamin D_3 have additive bone mineralization effects with bisphosphonates, selective estrogen-receptor modulators, or hormone therapies (e.g., estrogen for women and testosterone for men)? Is menaquinone more efficacious in reducing fracture risk than phytonadione? Does vitamin K play a role in immune modulation with respect to cytokine expression? Is there a potential relationship between chronic inflammation (as seen in both vascular calcification and osteoporosis) and vitamin K deficiency? For example, is synthesis or the activities of inflammatory cytokines, such as tumor necrosis factor, prostaglandin E_2, and interleukin-1, affected by vitamin K levels?

Because of their very low toxicity and potentially beneficial effects on both bone mineralization and attenuation of arterial calcification, phytonadione and menaquinone should be strongly considered as nutritional adjuncts in patients most susceptible to these disorders, such as postmenopausal women, diabetics, and hemodialysis patients.

Conclusion

Phytonadione and menaquinone

may be effective for the prevention and treatment of osteoporosis and arterial calcification.

References

1. Weber P. Vitamin K and bone health. *Nutrition.* 2001; 17:880-7.
2. Melton LJ III. How many women have osteoporosis now? *J Bone Miner Res.* 1995; 10:175.
3. Wallin R, Wajih N, Greenwood GT et al. Arterial calcification: a review of mechanisms, animal models, and the prospects for therapy. *Med Res Rev.* 2001; 21:274-301.
4. Arad Y, Spadaro LA, Goodman K et al. Predictive value of electron beam computed tomography of the coronary arteries: 19-month follow-up of 1173 asymptomatic subjects. *Circulation.* 1996; 93:1951-3.
5. Barengolts EI, Berman M, Kukreja SC et al. Osteoporosis and coronary atherosclerosis in asymptomatic postmenopausal women. *Calcif Tissue Int.* 1998; 62:209-13.
6. Hak AE, Pols HA, van Hemert AM et al. Progression of aortic calcification is associated with metacarpal bone loss during menopause: a population-based longitudinal study. *Arterioscler Thromb Vasc Biol.* 2000; 20:1926-31.
7. Jie KS, Bots ML, Vermeer C et al. Vitamin K intake and osteocalcin levels in women with and without aortic atherosclerosis: a population-based study. *Atherosclerosis.* 1995; 116:117-23.
8. Jie KS, Bots ML, Vermeer C et al. Vitamin K status and bone mass in women with and without aortic atherosclerosis: a population-based study. *Calcif Tissue Int.* 1996; 59:352-6.
9. Matsunaga S, Ito H, Sakou T. The effect of vitamin K and D supplementation on ovariectomy-induced bone loss. *Calcif Tissue Int.* 1999; 65:285-9.
10. Hauschka PV, Carr SA. Calcium-dependent α-helical structure in osteocalcin. *Biochemistry.* 1982; 21:2538-47.
11. Price PA. Gla-containing proteins of bone. *Connect Tissue Res.* 1989; 21:51-60.
12. Iwamoto J, Takeda T, Ichimura S. Effect of combined administration of vitamin D3 and vitamin K2 on bone mineral density of the lumbar spine in postmenopausal women with osteoporosis. *J Orthop Sci.* 2000; 5:546-51.
13. Price PA. Vitamin K-dependent formation of bone Gla protein (osteocalcin) and its function. *Vitam Horm.* 1985; 42:65-108.
14. Luo G, Ducy P, McKee MD et al. Spontaneous calcification of arteries and cartilage in mice lacking matrix GLA protein. *Nature.* 1997; 386:78-81.
15. Yagami K, Suh JY, Enomoto-Iwamoto M et al. Matrix GLA protein is a developmental regulator of chondrocyte mineralization and, when constitutively expressed, blocks endochondral and intramembranous ossification in the limb. *J Cell Biol.* 1999; 147:1097-108.
16. Braam LA, Dissel P, Gijsbers BL et al. Assay for human matrix Gla protein in serum. *Arterioscler Thromb Vasc Biol.* 2000; 20:1257-61.
17. Shanahan CM, Proudfoot D, Farzaneh-Far A et al. The role of Gla proteins in vascular calcification. *Crit Rev Eukaryot Gene Expr.* 1998; 8:357-75.
18. Price PA, Faus SA, Williamson MK. Warfarin causes rapid calcification of the elastic lamellae in rat arteries and heart valves. *Arterioscler Thromb Vasc Biol.* 1998; 18:1400-7.
19. Vermeer C, Gijsbers BL, Craciun AM et al. Effects of vitamin K on bone mass and bone metabolism. *J Nutr.* 1996; 126(4 suppl):1187S-91S.
20. Schurgers LJ, Vermeer C. Differential lipoprotein transport pathways of K-vitamins in healthy subjects. *Biochim Biophys Acta.* 2002; 1570:27-32.
21. Vermeer C, Braam L. Role of K vitamins in the regulation of tissue calcification. *J Bone Miner Metab.* 2001; 19:201-6.
22. Lipsky JJ. Nutritional sources of vitamin K. *Mayo Clin Proc.* 1994; 69:462-6.
23. Suttie JW. The importance of menaquinones in human nutrition. *Annu Rev Nutr.* 1995; 15:399-417.
24. Berkner KL. The vitamin K-dependent carboxylase. *J Nutr.* 2000; 130:1877-80.
25. Sunnerhagen M, Drakenberg T, Forsen S et al. Effect of Ca^{2+} on the structure of vitamin K-dependent coagulation factors. *Haemostasis.* 1996; 26:45-53.
26. Vermeer C. Gamma-carboxyglutamate-containing proteins and the vitamin K-dependent carboxylase. *Biochem J.* 1990; 266:625-36.
27. Suttie JW. Synthesis of vitamin K-dependent proteins. *FASEB J.* 1993; 7:445-52.
28. Furie B, Bouchard BA, Furie BC. Vitamin K-dependent biosynthesis of γ-carboxyglutamic acid. *Blood.* 1999; 93:1798-808.
29. Binkley NC, Krueger DC, Engelke JA et al. Vitamin K supplementation reduces serum concentrations of under-gamma-carboxylated osteocalcin in healthy young and elderly adults. *Am J Clin Nutr.* 2000; 72:1523-8.
30. Sokoll LJ, Sadowski JA. Comparison of biochemical indexes for assessing vitamin K nutritional status in a healthy adult population. *Am J Clin Nutr.* 1996; 63:566-73.
31. Sokoll LJ, Booth SL, O'Brien ME et al. Changes in serum osteocalcin, plasma phylloquinone, and urinary γ-carboxyglutamic acid in response to altered intakes of dietary phylloquinone in human subjects. *Am J Clin Nutr.* 1997; 65:779-84.
32. Binkley NC, Krueger DC, Kawahara TN et al. A high phylloquinone intake is required to achieve maximal osteocalcin gamma-carboxylation. *Am J Clin Nutr.* 2002; 76:1055-60.
33. Booth SL, Martini L, Peterson JW et al. Dietary phylloquinone depletion and repletion in older women. *J Nutr.* 2003; 133:2565-9.
34. Koshihara Y, Hoshi K, Ishibashi H et al. Vitamin K2 promotes 1-alpha-25(OH)$_2$ vitamin D3-induced mineralization in human periosteal osteoblasts. *Calcif Tissue Int.* 1996; 59:466-73.
35. Koshihara Y, Hoshi K, Okawara R et al. Vitamin K stimulates osteoblastogenesis and inhibits osteoclastogenesis in human bone marrow cell culture. *J Endocrinol.* 2003; 176:339-48.
36. Braam LA, Knapen MH, Geusens P et al. Vitamin K1 supplementation retards bone loss in postmenopausal women between 50 and 60 years of age. *Calcif Tissue Int.* 2003; 73:21-6
37. Hirano J, Ishii Y. Effects of vitamin K$_2$, vitamin D, and calcium on the bone metabolism of rats in the growth phase. *J Orthop Sci.* 2002; 7:364-9.
38. Feskanish D, Weber P, Willett WC et al. Vitamin K intake and hip fractures in women: a prospective study. *Am J Clin Nutr.* 1999; 69:74-9.
39. Booth SL, Tucker KL, Chen H et al. Dietary vitamin K intakes are associated with hip fracture but not with bone mineral density in elderly men and women. *Am J Clin Nutr.* 2000; 71:1201-8.
40. Vergnaud P, Garnero P, Meunier PJ et al. Undercarboxylated osteocalcin measured with a specific immunoassay predicts hip fracture in elderly women: the EPIDOS study. *J Clin Endocrinol Metab.* 1997; 82:719-24.
41. Szulc P, Chapuy MC, Meunier PJ et al. Serum undercarboxylated osteocalcin is a marker of the risk of hip fracture in elderly women. *J Clin Invest.* 1993; 91:1769-74.
42. Szulc P, Chapuy MC, Meunier PJ et al. Serum undercarboxylated osteocalcin is a marker of the risk of hip fracture: a three year follow-up study. *Bone.* 1996; 18:487-8.
43. Knapen MH, Nieuwenhuijzen Kruseman AC, Wouters RS et al. Correlation of serum osteocalcin fractions with bone mineral density in women during the first 10 years after menopause. *Calcif Tissue Int.* 1998; 63:375-9.
44. Schaafsma A, Muskiet FA, Storm H et al. Vitamin D$_3$ and vitamin K$_1$ supplementation of Dutch postmenopausal women with normal and low bone mineral densities: effects on serum 25-hydroxyvitamin D and carboxylated osteocalcin. *Eur J Clin Nutr.* 2000; 54:626-31.
45. Hodges SJ, Pilkington MJ, Stamp TC et al. Depressed levels of circulating menaquinones in patients with osteoporotic fractures of the spine and femoral neck. *Bone.* 1991; 12:387-9.
46. Shiraki M, Shiraki Y, Aoki C et al. Vitamin K$_2$ (menatetrenone) effectively prevents fractures and sustains lumbar bone mineral density in osteoporosis. *J Bone Miner Res.* 2000; 15:515-21.
47. Miki T, Nakatsuka K, Naka H et al. Vitamin K2 (menaquinone 4) reduces serum undercarboxylated osteocalcin level as early as 2 weeks in elderly women with

established osteoporosis. *J Bone Miner Metab.* 2003; 21:161-5.

48. Douglas AS, Robins SP, Hutchison JD et al. Carboxylation of osteocalcin in postmenopausal osteoporotic women following vitamin K and D supplementation. *Bone.* 1995; 17:15-20.
49. Takahashi M, Naitou K, Ohishi T et al. Effect of vitamin K and/or D on undercarboxylated and intact osteocalcin in osteoporotic patients with vertebral or hip fractures. *Clin Endocrinol.* 2001; 54:219-24.
50. Iwamoto J, Takeda T, Ichimura S. Treatment with vitamin D3 and/or vitamin K2 for postmenopausal osteoporosis. *Keio J Med.* 2003; 52:147-50.
51. Beresford JN, Gallagher JA, Poser JW et al. Production of osteocalcin by human bone cells in vitro: effect of 1,25 $(OH)_2 D_3$ 24, 25 $(OH)_2 D_3$, parathyroid hormone, and glucocorticoids. *Metab Bone Dis Relat Res.* 1984; 5:229-34.
52. Caniggia A, Nuti R, Galli M et al. Effect of a long-term treatment with 1,25-dihydroxyvitamin D_3 on osteocalcin in postmenopausal osteoporosis. *Calcif Tissue Int.* 1986; 38:328-32.
53. Lian JB, Carnes DJ, Glimcher MJ. Bone and serum concentrations of osteocalcin as a function of 1,25-dihydroxyvitamin D_3 circulating levels in bone disorders in rats. *Endocrinology.* 1987; 120:2123-30.
54. Hidaka T, Hasegawa T, Fujimura M et al. Treatment for patients with postmenopausal osteoporosis who have been placed on HRT and show a decrease in bone mineral density: effects of concomitant administration of vitamin K2. *J Bone Miner Metab.* 2002; 20:235-9.
55. National Institutes of Health. Vitamin K and bone turnover in postmenopausal women. www.clinicaltrials.gov/ct/show/NCT00062595?order=11 (accessed 2004 Jun 21).
56. Dhore CR, Cleutjens JP, Lutgens E et al. Differential expression of bone matrix regulatory proteins in human atherosclerotic plaques. *Arterioscler Thromb Vasc Biol.* 2001; 21:1998-2003.
57. Doherty TM, Detrano RC. Coronary arterial calcification as an active process: a new perspective on an old problem. *Calcif Tissue Int.* 1994; 54:224-30.
58. Shanahan CM, Cary NR, Salisbury JR et al. Medial localization of mineralization-regulating proteins in association with Monckeberg's sclerosis: evidence for smooth muscle cell-mediated vascular calcification. *Circulation.* 1999; 100: 2168-76.
59. Stehbens WE. Atherosclerosis and degenerative diseases of blood vessels. In: Stehbens WE, Lie JT, eds. Vascular pathology. London: Chapman and Hall Medical; 1995.
60. Spronk HM, Soute BA, Schurgers LJ et al. Matrix Gla protein accumulates at the border of regions of calcification and normal tissue in the media of the arterial vessel wall. *Biochem Biophys Res Commun.* 2001; 289:485-90.
61. Farzaneh-Far A, Proudfoot D, Weissberg PL et al. Matrix Gla protein is regulated by a mechanism functionally related to the calcium-sensing receptor. *Biochem Biophys Res Commun.* 2000; 277:736-40.
62. Geleijnse JM, Vermeer C, Grobbee DL et al. Dietary intake of menaquinone is associated with a reduced risk of coronary heart disease: the Rotterdam study. *J Nutr*. 2004; 134:3100-5.
63. Braam LA, Hoeks AP, Brouns F et al. Beneficial effects of vitamins D and K on the elastic properties of the vessel wall in postmenopausal women: a follow-up study. *Thromb Haemost.* 2004; 91:373-80.
64. Vitamin K monograph. Natural Medicines Comprehensive Database.
65. Vitamin K monograph. Micromedex Healthcare Series. Englewood, CO: Micromedex Inc.
66. McEvoy GK, ed. AHFS drug information. Bethesda, MD: American Society of Health-System Pharmacists; 1998.