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Vitamin K2 as MK-7: A Critical Nutrient for Better Osteoporosis Outcomes

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In this paper, we contrast the two contributors of bone strength defined by the National Institutes of Health (NIH): bone quantity and bone quality. Bone quantity, measured as bone mineral density (BMD), is a static measure that, while valuable, only gives insight into one aspect of bone. We, therefore, argue that bone quantity as BMD alone is far too simplified as the gold standard metric of bone strength. Instead, we favor emphasizing the more comprehensive measure, bone quality, and even further – bone dynamics – which goes beyond bone microarchitecture and considers the functional role of bone metabolism and the metabolic reserve of the bone in overall bone strength. We also detail the role of MK-7, a vitamin K2 species, for its emerging role in supporting practical measures of bone dynamics. We contrast the relative superiority of MK-7 to other commercially available forms of vitamin K in terms of bioavailability, serum half-life, and its ability to carboxylate the important bone matrix protein, osteocalcin (OC). We detail the mechanisms underlying MK-7's role in bone metabolism through the carboxylation of osteocalcin and the beneficial and suppressive effects of MK-7 on osteoblasts and osteoclasts, respectively. The potential role of vitamin K in bisphosphonate therapy and the beneficial role vitamin K has on regulating the inhibition of arterial calcification is discussed. Finally, we give clinical recommendations for assessing bone strength considering bone dynamics and bone density and the advantage of utilizing a synergistic combination of micronutrients for optimal bone strength.

Introduction

Although bone is classically regarded for its roles in skeletal structure, locomotion, protection of internal organs, and regulation of mineral homeostasis, bone is increasingly recognized as a metabolically active tissue with diverse physiological functions.[1,2] Bone constantly undergoes a back-and-forth remodeling process mediated by osteoblasts and osteoclasts to replace old bone with new bone, a process that begins before birth and continues until death.[3,4] In the context of bone health, the day-to-day physiological resilience of bone is either supported or depleted depending upon one's day-to-day internal inputs and lifestyle signals (e.g., nutrient status, hormone levels, age, activity levels, pharmaceutical use, inflammation, and other insults such as cigarette smoking and alcohol use). For instance, the presence of bone-building nutrients in the daily diet (i.e., vitamins K, D, calcium, magnesium) supports the day-to-day bone remodeling process. When compounded over time, like a long-term investment, a large buffer or safety net is created – referred to in the case of lifestyle medicine as metabolic reserve.[†] The long-term metabolic reserve of the bone, is then the result of lifestyle factors (good or bad) compounded over time. The importance of maintaining a strong bone metabolic reserve is magnified when a large stress or insult takes place.

A myriad of signals in our modern lives stress the physiological resilience and eventually the metabolic reserve of the bone, often with a multifactorial etiology. Some lifestyle stressors on bone include the Standard American Diet, cigarette smoking, sedentary activity patterns, and chronic inflammatory conditions.[4] For example, some subjects with inflammatory bowel disease (IBD) experience extraintestinal manifestations including metabolic bone disease (e.g., osteopenia and osteoporosis).[5] A range of internal host factors and lifestyle signals compromise the physiological resilience of bone in IBD including inflammation-related bone resorption, dietary malabsorption of minerals, vitamin D deficiency, alterations in the bone-immune signaling interaction, and even secondary effects from the pharmacological steroid therapy whose aim is to keep the immune system in a quiescent state.[5] These, amongst other factors, act together or individually to affect the bone's metabolic reserve. IBD is just one example of many inflammatory conditions that may affect bone strength.

[†] For detailed information on the importance of physiological resilience and metabolic reserve in the context of lifestyle-based therapies, see "The Standard" Road Map Series, which can be accessed at LifestyleMatrix.com.

It is important to note that the factors affecting the bone's physiological resilience and metabolic reserve (e.g., genetics, health status, lifestyle experiences, etc.) are extremely individual and unique to each patient. Positive lifestyle changes, no matter how small, sustained over a long period of time, are the most powerful way to rebuild metabolic reserve and regain physiologic resilience. These progressive changes provide a strong foundation and enhance additional therapeutic agents. Below we detail the intricate mechanisms by which vitamin K2 as MK-7 supports bone strength and can be leveraged to help build bone's physiological resilience and metabolic reserve. Because detailing the mechanisms of all nutrients and lifestyle factors beneficial to bone health are beyond the scope of this review, we recognize here that MK-7 is one important lifestyle signal that fits into a comprehensive micronutrient and lifestyle therapeutic plan for bone health.

Quality and Quantity as It Applies to Bones

In 2000, the NIH Consensus Development Panel defined osteoporosis as a *"skeletal disease characterized by decreased **bone strength** and associated with increased risk of fractures."* The Panel explained that bone strength has two components: **bone density (quantity)** and **bone quality**.[6] Bone quantity or density is expressed as grams of mineral per area of volume typically measured via a noninvasive DEXA scan, and is currently the gold standard diagnostic tool in osteopenia and osteoporosis. However, research has highlighted some shortcomings of BMD for predicting fracture risk. For example, analysis of data from the Rotterdam Study (N = 7,806), a large population-based study of diseases in the elderly, found that only 44% of women and 21% of men with non-vertebral fracture had BMD in the osteoporotic range (T-score below -2.5), suggesting that more sensitive risk assessment tools are needed.[7]

As the NIH explains, there is another component to bone strength, termed **bone quality**. Bone quality considers many additional structural aspects of bone, such as bone microarchitecture, bone turnover, bone damage accumulation (e.g., microfractures), bone mineralization (or calcification), bone matrix composition (including collagen structure), and bone mineral composition.[8] Although historically, quantifying bone quality was difficult and invasive, more recent technology is emerging to quantify changes to bone quality such as measuring bone turnover biomarkers and imaging methods for visualizing bone microarchitecture and bone matrix composition.[8,9] Despite bone quality being more comprehensive than bone quantity, bone quality has been criticized for still being too focused on the physical structure of bone. In fact, bone quality was proposed as a term when BMD measures alone could not explain fracture risk reduction in multicenter clinical trials using two classes of pharmaceutical drugs (i.e., antiresorptive bisphosphonate therapy and selective estrogen receptor modulator therapy).[9] Therefore, bone quality still lacks emphasis on bone metabolism and metabolic reserve. In response to these data, we use the term **bone dynamics** to build on bone quality by further emphasizing the dynamics of bone metabolism that govern bone protein production, the metabolic reserve that balances bone remodeling, and key nutrients that contribute to both. For our purposes, we use the phrase **bone dynamics** to expand upon bone quality in our review.

We define bone dynamics as the sum of the structural and material properties of the bone, the dynamic balance of bone metabolism, and the metabolic reserve of foundational bone nutrients.[10] Consequently, we argue that measuring bone dynamics along with traditional bone mineral density provides a multifaceted measure that includes both metabolic and microstructural indices of the bone and a more accurate reflection of overall bone strength compared to measuring BMD alone. Specific biomarkers are addressed further in the paper. In fact, a 2018 study highlighted the relative superiority of measuring bone quality, a key aspect of bone dynamics, compared to solely relying on bone mineral density measures for bone strength outcomes. More specifically, better correlation was found between bone strength and component measures of bone dynamics measured via histomorphometry (e.g., bone volume, trabecula number, trabecular separation, and trabecular thickness) compared to bone mineral density when analyzing cubical bone specimens obtained from patients undergoing hip replacement procedures.[10,7] Based off these data, we argue that bone dynamics are an underrepresented and perhaps, much needed, missing piece in the bone strength puzzle compared to bone mineral density alone.[†] Therefore, the translation of the bone dynamics into research and clinical practice is needed to better predict

[†] Although we argue that measures of bone dynamics may give more insight to the metabolic reserve of the bone than BMD measures alone, we acknowledge that research and clinical practice heavily rely on measures of bone mineral density when evaluating bone outcomes in clinical trials and in the clinic. Therefore, we argue for additional research on bone dynamics outcomes, and argue for their application to supplement BMD measures for additional information on the metabolic reserve of the bone.

fracture risk and to understand the mechanisms that micronutrients and pharmacological interventions have on bone dynamics and consequently, bone strength.[8]

Metabolic Aspects of Bone Dynamics

Emerging research suggests vitamin K2 may improve measures of bone quality and improve many metabolic aspects of bone dynamics. One animal study found the addition of vitamin K2 to bisphosphate therapy ameliorated the suppressive effect of bisphosphonates on bone turnover, increased bone volume, and increased bone formation parameters.[11] Another study evaluated the synergistic ability of vitamin K2 and vitamin D3 to improve measures of bone quality in osteoblasts from obese diabetic mice.[12] When treating osteoblasts from these animals with vitamin K2 and vitamin D3, many metabolic measures of bone dynamics improved. More specifically, enhanced calcium deposition in bone was found, levels of the bone matrix protein osteocalcin increased, markers associated with bone formation increased (e.g., alkaline phosphatase activities [ALP]), bone formation transcription factors), and improved bone microarchitecture was found. These animal studies serve as a model for compromised bone dynamics at baseline and shed light on key nutrients that should be considered during assessment and for therapeutic benefit. Further, these mechanistic studies suggest that vitamin K may improve a diverse array of bone dynamics measures, which is a necessary component to consider when assessing bone strength. Clinically, measuring the status of vitamins D and K via laboratory analysis may also serve as key markers of bone dynamics – see sidebar below for our recommendation for assessing bone dynamics in the clinic.

Recommendation for Assessing Bone Dynamics

Although the most common measure of bone strength currently is bone mineral density via DEXA scan, a measure of bone quantity, this measure only considers one static measure of bone strength. Bone mineral density is commonly used because it is noninvasive and easy to measure. However, emerging biomarkers of bone metabolism, bone quality, and nutrient status are becoming available for the clinician. Here we detail some emerging biomarkers.

Undercarboxylated osteocalcin (ucOC), as discussed throughout this review, is a functional test that gives the clinician a picture of the patient's vitamin K status and function as it relates to bone health. We recommend that ucOC should be measured along with a DEXA scan to get a more accurate picture of bone dynamics and overall bone strength.[13] Undercarboxylated osteocalcin is available as an assay at commercial laboratories.

Bone formation markers such as alkaline phosphatase (ALP) or osteocalcin. Increased alkaline phosphatase activity is indicative of active bone formation and is a byproduct of osteoblast activity.

Collagen degradation markers are indicators of bone resorption. These markers include C-terminal telopeptide of type I collagen (CTx I), pyridinoline cross-links (PYP) or deoxypyridinoline (DPD). These measures are valuable in gauging treatment efficacy and ensure measures of bone dynamics are considered.[13]

Vitamin D status, as vitamin D is an important nutrient for many aspects of health, including bone health. Vitamin D (amongst other factors) has been shown to influence the osteoblastic synthesis of osteocalcin, and has, in some studies, been shown to decrease undercarboxylated levels of osteocalcin.[14,15] Therefore, we recommend measuring serum levels of vitamin D as 25-hydroxyvitamin D to estimate the metabolic reserve capacity of this important nutrient for bone health.

Vitamin K as an Essential Bone Strength Nutrient

Although vitamin K is perhaps best known for its role in blood coagulation as a necessary cofactor in the carboxylation of hepatic blood clotting factors,[†] vitamin K also participates in the carboxylation of extra-hepatic vitamin-K dependent proteins (VKD) important for a range of physiological functions such as proper calcium deposition in bones (via the VKD protein osteocalcin) and preventing arterial calcification (via the VKD protein matrix Gla protein).[19,20] Vitamin K exists in two major forms: vitamin K1 (or phylloquinone) and vitamin K2 (or menaquinone). Both forms have the same ring structure, called menadione (the active site for vitamin K), but are differentiated structurally by the saturation and length of their side chains and their dietary sources.[21]

Phylloquinone (vitamin K1) is the most common dietary form of vitamin K, synthesized by plants and cyanobacteria, and is found in green leafy vegetables and vegetable oils.[22] Whereas menaquinones (vitamin K2) are synthesized by certain bacteria which produce repeating prenyl units as a sidechain, the number of prenyl sidechains gives rise to the classification of menaquinones (MK-2 to MK-14). Menaquinones are present in the diet mostly in fermented foods (e.g., natto, cheese, etc.) or by bacteria present in some foods (e.g., liver).[22,23] Microbes in the human gut also synthesize menaquinones, but their contribution to host vitamin K status is unknown.[24,25,26,27] Both phylloquinone and menaquinones have similar biological activities with some notable differences in their pharmacokinetics and their receptor binding affinities due to the differences in their sidechains.[28,29]

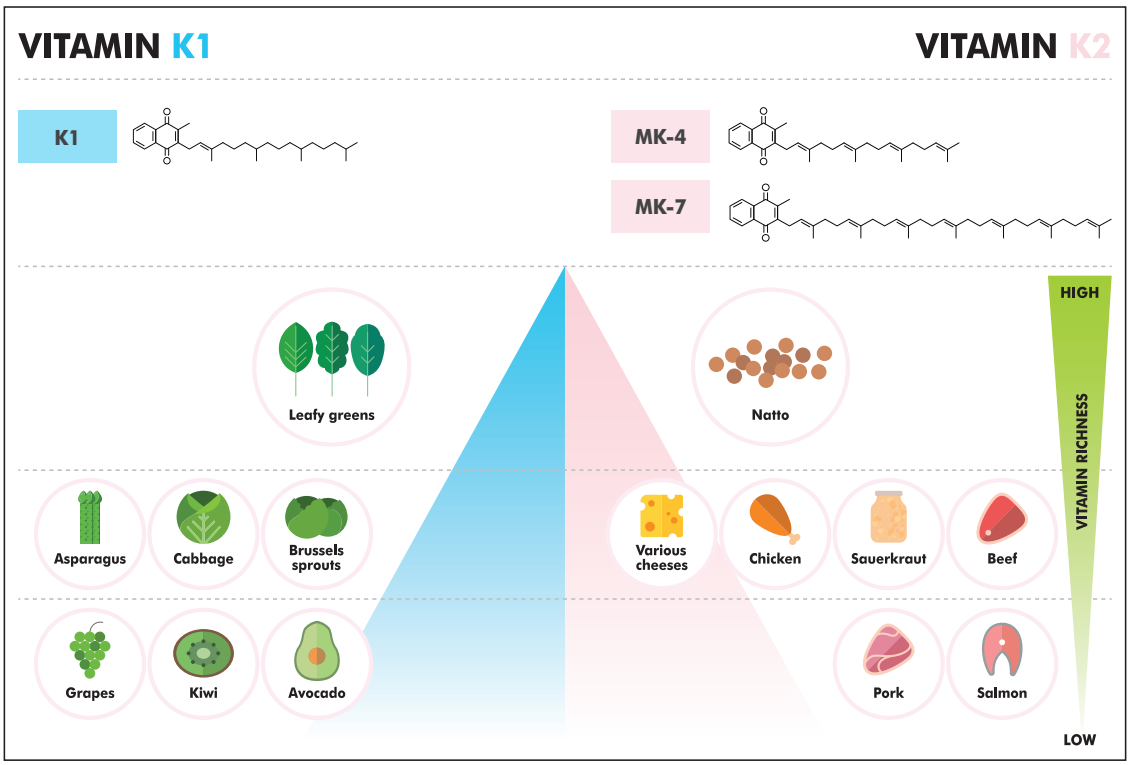

Figure 1. Primary sources and chemical structure of vitamin K1 and vitamin K2. Image adapted from Halder M, Petsophonsakul P, Akbulut AC, et al. Vitamin K: Double Bonds beyond Coagulation Insights into Differences between Vitamin K1 and K2 in Health and Disease. *Int J Mol Sci.* 2019;20(4). This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).

[†] Although the interaction of vitamin K with vitamin K antagonist therapy is beyond the scope of this review, studies have shown vitamin K may affect coagulation status in those taking VKA therapy.[16,17] Of note, an intervention study found that MK-7 in doses up to 360 µg/day for 12 weeks did not affect coagulation status in healthy subjects not taking VKA therapy, so this caution is only for those taking VKA therapy.[18] As with all therapies, clinicians should consider any drug-nutrient interactions before administering a therapy. Clinicians should monitor dietary and supplemental vitamin K intake along with monitoring INR status in those taking VKA therapy.

Comparison of Vitamin K Supplement Forms: The Superiority of Vitamin K2 as MK-7

It is important for clinicians to understand which vitamin K forms are commercially available as dietary supplements and to understand how these forms compare in terms of bioavailability, half-life, and their relative ability to carboxylate osteocalcin. Vitamin K1, or phylloquinone, is the form most used in supplements as a "bioidentical" compound produced via organic synthesis and is usually given in dosages of one mg per day.[30,31] Although there are many vitamin K2 or menaquinone compounds present in nature, only two are available as dietary supplements (i.e., MK-4 and MK-7). It is important to note that not only do the two available menaquinones differ in their side-chain length, the MK-4 and MK-7 forms also differ considerably in their dosages by orders of magnitude. Due to the greater bioavailability of MK-7 relative to MK-4, the clinically relevant dosage for MK-4 is magnitudes higher ~45 **mg/day**, compared to only 45–360 **µg/day** for MK-7.[32,†] Overall, studies have shown the relative superiority of MK-7 over the other forms of vitamin K.

Bioavailability and Half-life

A human pharmacokinetic study compared oral supplementation with MK-7 head-to-head with phylloquinone *(see Figure 2)* and found that although both were well-absorbed and both peaked in the serum after four hours of supplementation, MK-7 accumulated to much higher serum levels (seven- to eight-fold higher), had a much longer serum half-life, and reached more stable serum levels with prolonged intake.[16] These results suggest that MK-7, due to its longer serum half-life compared to phylloquinone, may be available in the tissue longer for extrahepatic tissue uptake and; subsequently, greater availability for more complete carboxylation of osteocalcin. It has been shown that the higher menaquinones (e.g., MK-7) are more hydrophobic; thus, are characterized by longer half-life times *in vivo*.[33]

Carboxylation and NF-kB Modulation

Studies indicate that a smaller dose of MK-7 can γ-carboxylate osteocalcin compared to doses of vitamin K1 or MK-4.[34] Further, natto-derived MK-7 has been shown to more completely carboxylate osteocalcin compared to vitamin K1.[16]

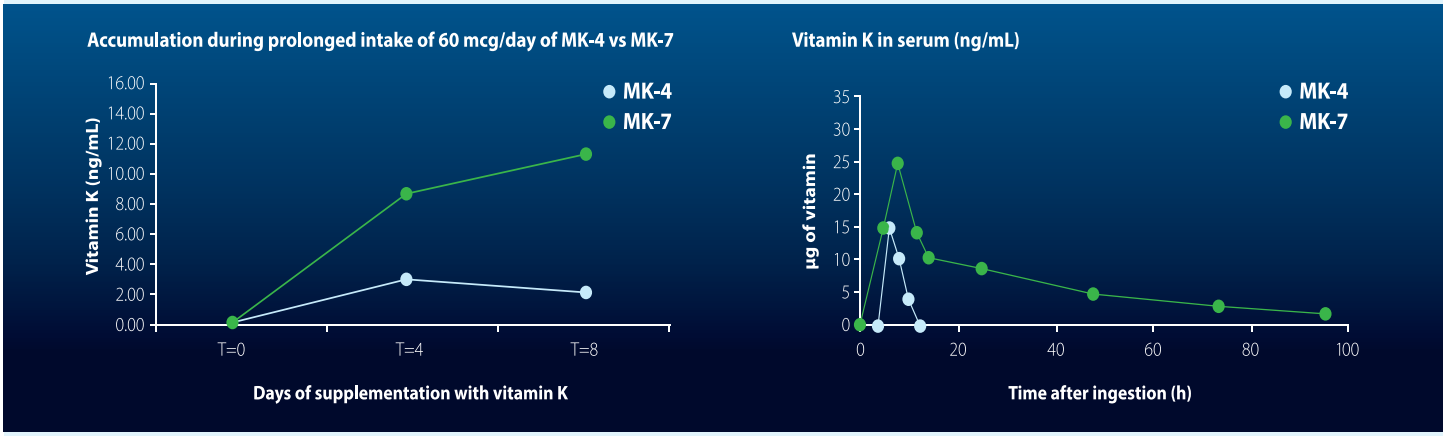

Figure 2. Bioavailability comparison between MK-4 and MK-7. Image adapted with permission from Schurgers LJ, Teunissen KJ, Hamulyak K, Knapen MH, Vik H, Vermeer C. Vitamin K-containing dietary supplements: comparison of synthetic vitamin K1 and nattoderived menaquinone-7. *Blood.* 2007;109(8):3279-3283.

† For additional information on vitamin K and dosage forms, the interested reader is directed to the following book, *Supplementing Dietary Nutrients: A Guide for Healthcare Professionals, Second Edition* (The Point Institute, 2020) which can be accessed from LifestyleMatrix.com.

Vitamin K2 has been shown to suppress basal and cytokine-induced NF-kB activation in a γ-carboxylation independent manner.[35] Suppressing NF-kB activation in this context has beneficial effects on bone metabolism by stimulating osteoblastogenesis and suppressing osteoclastogenesis.

Conversion of MK-7 to MK-4

Although MK-7 appears to more completely carboxylate OC, one *in vitro* study did find that MK-4 was able to induce expression of two genes in osteoblasts that MK-7 was not able to induce (i.e., growth differentiation factor and stanniocalcin); however, it should be noted that MK-7 is transformed to MK-4 in target tissues.[36,37] In fact, all vitamin K homologs are converted to MK-4 in tissues.[37,38] MK-4 is considered to perform other specific functions other than γ-carboxylation of VKDPs, such as modulation of the steroid and xenobiotic receptor SXR.[39] Ironically, a review paper suggests that MK-4 supplementation in rats may not raise extrahepatic MK-4 tissue levels whereas supplementation of MK-7 may significantly raise extrahepatic tissue MK-4 levels.[40] Additional research is needed to better understand this relationship, especially in terms of dosing. This could be partly explained by MK-4's poor bioavailability relative to MK-7 *(see Comparison of Vitamin K Supplement Forms)*.[32]

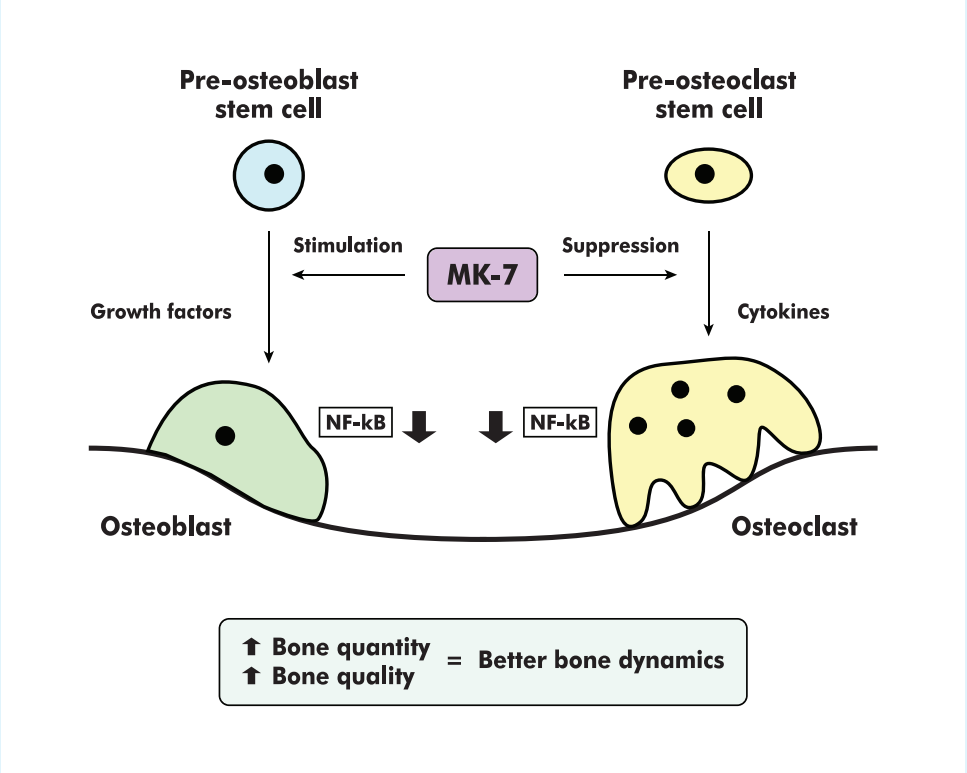

Figure 3. Cellular mechanisms by which menaquinone-7 (MK-7) stimulates osteoblastic bone formation and suppresses osteoclastic bone resorption. MK-7 suppresses NK-κB signaling pathways, which are activated by stimulation of TNF-α or RANKL, in osteoblasts and osteoclasts. In addition, MK-7 stimulates protein synthesis (including osteocalcin and other protein molecules) in osteoblastic cells, and it may regulate the gene expression of various proteins, which are related to cellular functions in osteoblastic and osteoclastic cells.

Image adapted from Yamaguchi M, Weitzmann MN. Vitamin K2 stimulates osteoblastogenesis and suppresses osteoclastogenesis by suppressing NF-κB activation. *Int J Mol Med.* 2011;27(1):3-14.

The Role of MK-7 in Bone Dynamics

The complete adult skeleton is replaced every decade in humans. This is accomplished by the metabolically active bone which contains four major types of bone cells: osteoclasts, osteoblasts, osteocytes and bone-lining cells. These cells interact in a coordinated manner to actively renew the skeleton. Recently, attention on vitamin K has shifted from its more widely known role in blood coagulation to its role in bone metabolism as well as its protective capacity in cardiovascular health as more research has emerged surrounding the role of vitamin K-dependent proteins in these processes (i.e., osteocalcin and matrix Gla protein). Because of the connection between vitamin K and the important bone protein, osteocalcin, vitamin K has been studied for its role in bone health in epidemiological and intervention studies. *In vitro* studies suggest vitamin K participates in bone metabolism via mechanisms independent of its gamma-carboxylation effects as well.[41,35] Vitamin K's mechanisms on bone homeostasis are extensive involving several cells within bone including osteoblasts, osteoclasts and osteocytes as well as genes which impact overall bone function *(see Figure 4)*.

Figure 4. The mechanism of action of vitamin K2 (as MK-7) in bone health includes stimulating osteoblasts, inhibiting osteoclasts and enhancing bone microarchitecture and mineralization. Image adapted from Akbari S, Rasouli-Ghahroudi AA. Vitamin K and Bone Metabolism: A Review of the Latest Evidence in Preclinical Studies. *BioMed Research International.* 2018;2018:4629383-4629383.

MK-7 Effects on Osteocalcin Function

Vitamin K's relationship to bone health is perhaps best known for its activation of the vitamin K-dependent protein, osteocalcin (OC). Osteocalcin (OC) is synthesized by osteoblasts and is known primarily for its role in bone formation and mineralization.[42] In fact, osteocalcin is one of the most abundant non-collagen proteins in bone. Osteocalcin contains three gamma-carboxyglutamic acid residues that require carboxylation by vitamin K to bind free calcium and hydroxyapatite and eventually participate in bone mineralization.[43,42] The degree of osteocalcin carboxylation can be measured and has been used as a biomarker for bone metabolism, an important component of bone dynamics.[†] Vitamin K deficiency leads to an increase in serum undercarboxylated osteocalcin (ucOC), and elevated concentrations of circulating ucOC have been associated with low bone mineral density, increased risk for hip fractures, and recognized as an independent risk factor for fractures.[44,45,15]

[†] It is important to note that the **percentage of undercarboxylated osteocalcin (ucOC)** is influenced by vitamin K status, while the **total serum osteocalcin** is influenced by osteoblastic synthesis and is independent of vitamin K.

Intervention with MK-7 in humans has been shown to improve the carboxylation status of osteocalcin.[†] A randomized, double-blind, placebo-controlled, parallel group comparison study reported that 12 weeks of daily MK-7 intake at 100 µg improved osteocalcin γ-carboxylation status in healthy men and women between the ages of 20 and 69 years.[46] Compared to placebo (0 µg MK-7 for 12 weeks), the MK-7 group's levels of undercarboxylated osteocalcin decreased significantly at day 28 ($P < 0.05$). Additionally, the carboxylated osteocalcin/undercarboxylated osteocalcin ratio was significantly increased compared to placebo during the 12-week intake period ($P < 0.05$).[46] These findings suggest that an MK-7 intake of 100 µg per day in addition to vitamin K intake from the normal diet results in improved osteocalcin function which correlates with improved calcium deposition in bone and subsequently improved bone formation. These results are strengthened by earlier work[47] and reinforce the idea that greater intakes of vitamin K2 are beneficial for bone formation. For this reason, we suggest using ucOC as not only a measure of vitamin K status, but an important clinical measure of bone dynamics, and overall bone strength.

MK-7 Effects on Osteoblast Function

It is well established that vitamin K affects the proliferation and differentiation of osteoblasts through the γ-carboxylation pathway *(see Figure 5)*.[48] *In vitro* evidence suggest that treatment of osteoblasts with vitamin K2 results in increased alkaline phosphatase activity.[49,50] Increased alkaline phosphatase activity is indicative of active bone formation and is a byproduct of osteoblast activity. Additionally, vitamin K is important for the protection of osteoblasts against apoptosis.[51] Furthermore, vitamin K2 exerts its bone protective effects by upregulating osteogenic genes. For example, MK-7 has been reported to upregulate the steroid and xenobiotic receptor (SXR) in bone cells which subsequently increased expression of several osteoblast bone markers such as ALP, osteopontin, osteoprotegerin (OPG), and matrix Gla protein.[52,53]

Vitamin K2 Effects on Osteoclast Function

Not only is vitamin K important for bone formation by stimulating osteoblastogenesis but vitamin K is also important for bone metabolism by inhibiting osteoclastic bone resorption.[50] This inhibitory effect on bone resorption has been suggested to be due to vitamin K2's side chain.[54] In addition, vitamin K2 has been shown to induce osteoclast apoptosis *in vitro*.[55] Vitamin K2 also has the potential to stimulate expression of osteogenic genes which not only increases osteoblast differentiation and improves osteoblast overall function but also inhibits osteoclast function. For example, it was reported that OPG, a decoy protein that binds to the receptor activator of nuclear factor-KB ligand (RANKL) which inhibits osteoclast differentiation, increased (while RANKL decreased) in rats supplemented with vitamin K2.[56] Additionally, a bone resorption marker, type I collagen CTx, was shown

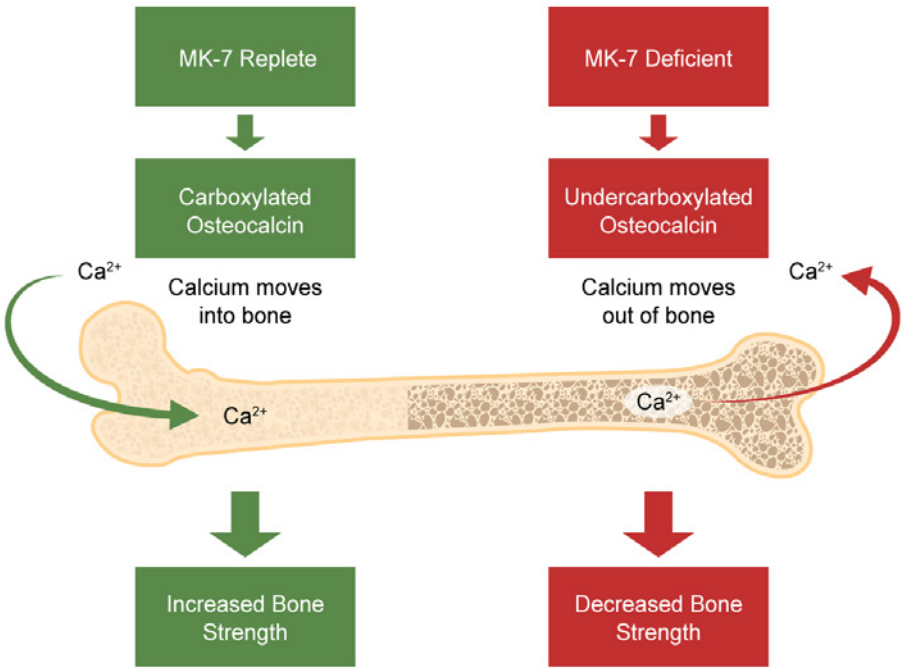

Figure 5. The effect of MK-7 on bone strength via the carboxylation of osteocalcin.

[†] Interestingly, there is an interconnection with vitamin D, as vitamin D (among other factors) has been shown to influence the osteoblastic synthesis of osteocalcin, and has, in some studies, been shown to decrease levels of undercarboxylated osteocalcin. The Role of vitamin D and OC is beyond the scope of this review, however, this relationship highlights nutrient synergy on bone health.[14,15]

to decrease when vitamin K2 was supplemented. Taken together, these results indicate that vitamin K2 supplementation positively influences bone metabolism.

MK-7 Enhances Collagen Production

As was discussed earlier, bone strength depends on both bone dynamics (e.g., shape of bones, microarchitecture of bone, bone turnover, mineral and collagen content, nutrient status, etc.) and bone quantity (e.g., density of bone). Collagen, in particular type I collagen, provides the flexible toughness to bone that enables it to absorb energy.[57] Compared to MK-4, MK-7 was shown to better increase collagen accumulation through osteoblastic cells.[58] These findings demonstrate the importance of vitamin K2 availability for quality bone formation.

Dietary MK-7 is Associated with Improved Bone Outcomes in Humans

Epidemiological studies suggest low intakes of vitamin K are associated with low bone mineral density and a higher risk of fracture.[59,60] Since MK-7 is limited to very few foods in the diet, most epidemiological studies have evaluated the MK-7 rich food, natto, which is a fermented soybean dish traditional to the Japanese diet. Natto contains 200-400 µg of MK-7 per serving of 30–45 g produced by *B. subtilis*.[34] Regional studies have shown that natto consumption is associated with a reduced incidence of hip fractures in women in Japan *(see Figure 6)*.[61,62]

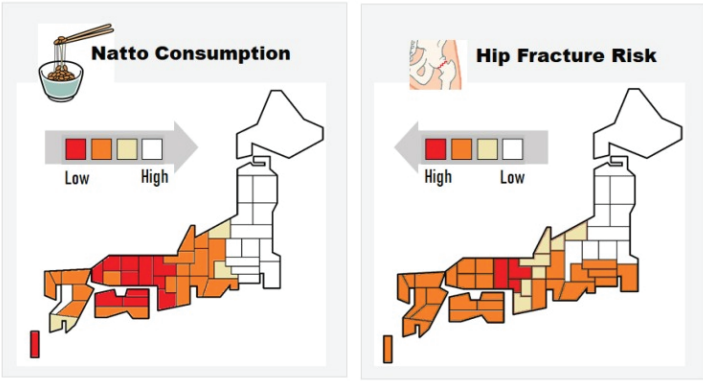

Figure 6. Comparison between regional natto consumption and the relative incidence of hip fracture in Japanese women.
Image adapted from Sato T, Inaba N, Yamashita T. MK-7 and Its Effects on Bone Quality and Strength. *Nutrients.* 2020;12(4):965. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (http://creativecommons.org/licenses/by/4.0/).

More specifically, increasing habitual natto intake has been associated with improved total hip BMD (P for trend = 0.0034) in healthy Japanese women (N = 944, 20-79 years) after a three-year follow-up period.[63]

Similarly, a cross-sectional study of 1,662 community-dwelling elderly Japanese men (≥ 65 years) found subjects with greater natto intakes (containing ≥ 350 µg/day MK-7) had significantly lower levels of serum ucOC, significantly higher BMD, and a lower risk of low BMD at the total hip and femoral neck compared to subjects consuming lower amounts of natto (containing < 50 µg/day of MK-7).[64]
A large prospective cohort study published in 2020 found similar results in that natto intake was inversely correlated with fracture risk in postmenopausal Japanese women (N = 1,417).[65] It appears that natto's unique properties, such as a high amount of MK-7, may underlie the benefit seen for fracture risk as the frequency of intake of other soybean products had no association with fracture risk. However, since epidemiological studies can only show correlation and cannot suggest causation, we look to human clinical trials for better understanding of the effect of MK-7 on bone health outcomes.

Human Clinical Trials Supplementing MK-7 for Bone Health

Although the efficacy of long-term MK-7 supplementation has received far less attention than that of MK-4 and phylloquinone, the available data on MK-7 for bone outcomes appears promising especially as the data on MK-4 and phylloquinone have been variable in outcomes.[21,66,4,67,68] One long-term, double-blind, randomized, placebo-controlled clinical trial found after three years of supplementing with 180 µg MK-7, healthy postmenopausal women (N = 244) had improved vitamin K status and a decreased age-related decline in BMD at the lumbar spine and femoral neck compared to placebo.[69] The improved vitamin K status was measured via a significant improvement in the ratio of undercarboxylated osteocalcin to carboxylated osteocalcin, which saw a maximal improvement after one year of supplementation and was maintained for the next two years. A significant improvement in bone mineral composition (BMC) and BMD of the femoral neck was not seen until year three of supplementation with MK-7, suggesting that other studies using relatively short vitamin K intervention periods (e.g., one year) may not have allowed sufficient time for the benefit on BMD and BMC of the femoral neck to be realized.[70] In contrast, significant improvements in BMC and BMD of the lumbar spine were seen after one year and two years, compared to placebo. Additional measures of bone strength were also significantly improved following MK-7 supplementation compared to placebo. MK-7 also prevented the loss in vertebral height of the lower thoracic region at the mid-site of the vertebrae.

The Postmenopausal Health Study II found a positive effect of MK-7 (100 µg/day) when given concomitantly with 800 mg calcium and 10 µg vitamin D3 (400 IU) on BMD to healthy postmenopausal women (N = 150) for twelve months via fortified milk and yogurt and lifestyle counseling.[71] Further, administering MK-7 with calcium and vitamin D was shown to significantly reduce levels of ucOC.

Clinical Considerations of MK-7

Bisphosphonate Treatment

Bisphosphonates are strong suppressors of osteoclasts and consequently slow the bone remodeling process, therefore increasing bone mineral density.[72] Because of this mechanism, bisphosphonates are commonly prescribed medications for osteoporosis; however, their use is not without potential adverse effects such as bisphosphonate-related osteonecrosis of the jaw after long-term use.[73,72] Bisphosphonates may also have negative effects on measures of bone dynamics such as reducing bone turnover and contributing to abnormal bone microstructure; therefore, an animal study evaluated the effect of combining bisphosphonate therapy with vitamin K2.[11] This study found that the addition of vitamin K2 to bisphosphate therapy could ameliorate the suppressive effect of bisphosphonates on bone turnover and increase the bone volume as well as the bone formation parameters, suggesting an improvement in measures of bone dynamics when given vitamin K2.[11]

Contributors to Arterial Calcification

Calcium supplementation is widely self-prescribed as a stand-alone treatment for bone loss. Several literature reviews suggest that stand alone calcium supplementation may offer limited benefit, along with questions regarding increasing risk of cardiovascular events.[74,75,76] Indeed, the data suggest that the rise in serum calcium from calcium supplementation may not only have positive effects on bone density but also, inadvertently, increase vascular calcification – precipitating cardiovascular events.[74,75] MK-7 supplementation in combination with lower doses of calcium may theoretically offer enhanced efficacy and safety given MK-7's ability to carboxylate another vitamin K-dependent protein produced by vascular smooth muscle cells and involved in the inhibition of arterial calcification, matrix Gla protein (a VKDP).[77,78,†] In this respect, vitamin K appears to have a dual role in regulating calcium deposition in that the VKDP osteocalcin directs calcium to bones, and the VKDP matrix Gla protein inhibits calcium deposition in the vasculature.

Although, to date, there are not many studies evaluating the concomitant supplementation of MK-7 with calcium for bone and arterial health outcomes, emerging research suggests giving nutrients concomitantly may provide synergistic benefit. For example, the Postmenopausal Health Study II described above found benefit for BMD and ucOC outcomes when combining MK-7 (100 µg/day) with 800 mg calcium and 10 µg vitamin D3 (400 IU) in 150 healthy postmenopausal women after twelve months.[71]

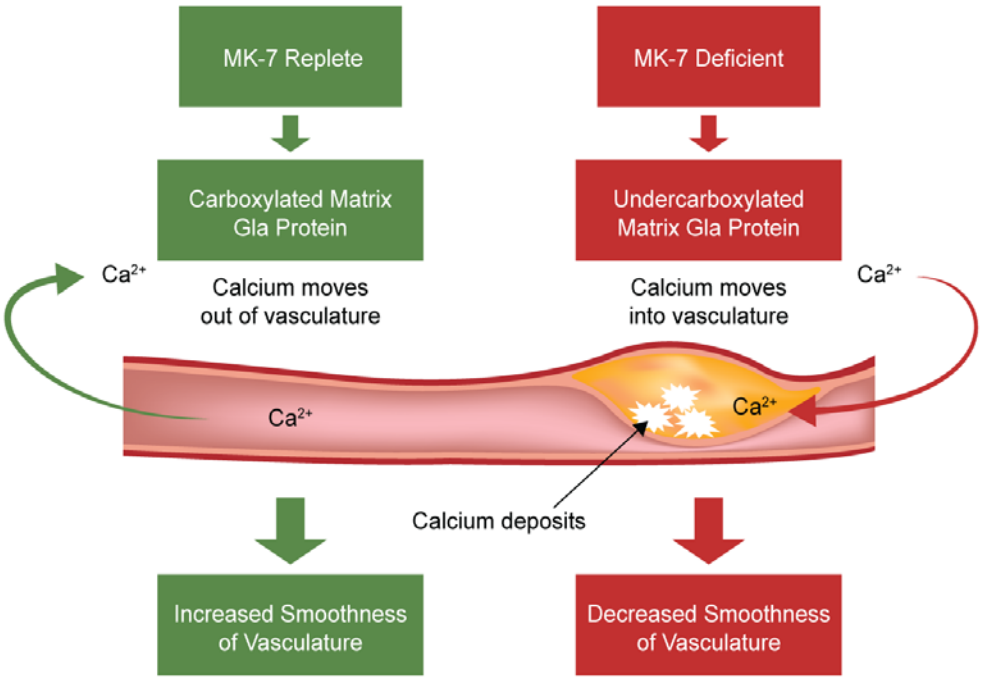

Figure 7. The effect of MK-7 on arterial calcification via the carboxylation of matrix Gla protein.

† For additional information on the relationship between vitamin K supplementation and cardiovascular outcomes, especially those related to inhibition of vascular calcification, the interested reader is directed to the following book, *Cardiometabolic Risk Management: A Functional and Lifestyle Approach*, which can be accessed from LifestyleMatrix.com.

We look forward to future human clinical trials studying the synergy of these nutrients for bone and arterial health outcomes. Other considerations should be made for those on corticosteroids due to the risk of secondary osteoporosis and statin drug use, as recent data has suggested an increase in arterial calcification with long-term use.[30]

Clinical Recommendations for MK-7 and Bone Strength

Although mechanism, epidemiological and human clinical trials show vitamin K2 as MK-7 is an important nutrient for bone strength, United States population data consistently reveal that the prevalence of inadequate vitamin K intake is high. In fact, NHANES data have shown that 58-69% of the US population does not meet adequate vitamin K intake levels from food alone.[79,80] Importantly, these NHANES data rely heavily on phylloquinone estimates as phylloquinone is the major dietary contributor to vitamin K status in U.S. populations. Outside of natto intake, MK-7 is very difficult to obtain from the diet. This reality is troubling given the relative superiority of MK-7 over other commercially available forms of vitamin K (i.e., phylloquinone and MK-4) in terms of bioavailability and bioactivity (especially, the carboxylation of osteocalcin). Therefore, it is important that clinicians consider the vitamin K status of their patients. When supplementation is necessary, we strongly recommend that MK-7 is given synergistically with other micronutrients indicated for bone health. These nutrients, when given concomitantly, have unique and often overlapping mechanisms that have been shown to compound to greater benefit.[71] More specifically, in order to maximize bone metabolic reserve and bone physiological resilience we recommend clinicians give calcium concomitantly with magnesium (150-300 mg/day from a combination of highly bioavailable forms such as organic salts and chelates), vitamin D3 (800-2,000 IU, depending on baseline levels), and vitamin K2 (45-180 µg menaquinone-7). Clinicians should recommend this supplementation protocol to any patient with a family history of osteoporosis, multiple primary and secondary risk factors for osteoporosis, patients approaching menopause and anyone 50 years of age and older to slow bone loss.

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