## Title

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## Authors

Shea, MK
Kritchevsky, SB
Hsu, F-C
et al.

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# The association between vitamin K status and knee osteoarthritis features in older adults: The Health, Aging and Body Composition Study

**M. Kyla Shea, PhD**[1], **Stephen B. Kritchevsky, PhD**[2], **Fang-Chi Hsu, PhD**[3], **Michael Nevitt, PhD**[4], **Sarah L. Booth, PhD**[1], **C. Kent Kwoh, MD**[5], **Timothy E. McAlindon, MD**[6], **Cees Vermeer, PhD**[7], **Nadja Drummen, BS**[7], **Tamara B. Harris, MD, MS**[8], **Catherine Womack, MD**[9], and **Richard F. Loeser, MD**[11] **for the Health ABC Study**

[1]USDA Human Nutrition Research Center on Aging at Tufts University, Boston MA [2]Sticht Center on Aging, Wake Forest School of Medicine, Winston-Salem NC [3]Department of Biostatistical Sciences, Division of Public Health Sciences, Wake Forest School of Medicine, Winston-Salem NC [4]Department of Epidemiology and Biostatistics, University of California San Francisco, San Francisco CA [5]Division of Rheumatology, University of Arizona, Tucson AZ [6]Division of Rheumatology, Tufts Medical Center, Boston, MA [7]VitaK, Maastricht, Netherlands [8]Laboratory of Epidemiology and Population Sciences, National Institute on Aging, Memphis, Tennessee [9]Department of Preventive Medicine, University of Tennessee Health Science Center, Memphis, Tennessee [11]Division of Rheumatology, Allergy and Immunology, University of North Carolina, Chapel Hill NC

**Keywords**

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Osteoarthritis (OA) is a debilitating joint-disease characterized by pathological changes in all joint tissues, including cartilage, bone, meniscus (in the knee), and synovium, causing joint pain and loss of function (1). Both cartilage and meniscal calcification have been implicated in OA (2;3). Because there is no known therapy to slow OA progression, the identification of simple and effective strategies that may reduce OA progression is important to delay disability and reduce the associated cost-burden.

Corresponding Author: Kyla Shea, PhD, USDA Human Nutrition Research Center on Aging at Tufts University, Boston MA; kyla.shea@tufts.edu.

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Vitamin K is a fat-soluble nutrient that appears to have a role in OA. In epidemiological studies, low circulating vitamin K was associated with greater prevalence of hand and knee OA cross-sectionally (4) and with greater knee OA progression and cartilage loss longitudinally (5). The main function of vitamin K is as an enzymatic cofactor for the gamma (γ)-carboxylation of certain calcium-binding proteins, including matrix gla protein (MGP), a vitamin K-dependent (VKD) mineralization inhibitor expressed in human articular cartilage (6). Once carboxylated, MGP inhibits ectopic mineralization by binding calcium crystals, thereby inhibiting calcium crystal growth, and by binding to and inhibiting bone morphogenic protein-2, a protein that induces bone formation (7–9). In human OA cartilage, MGP is primarily uncarboxylated, the less functional form), whereas in healthy articular cartilage MGP is primarily carboxylated (functional) (10), suggesting the carboxylation of MGP is relevant to OA. MGP is also detectable in circulation and desphospho-ucMGP [(dp)ucMGP] concentrations increase when vitamin K status is low (11), suggesting circulating (dp)ucMGP may serve as a functional biomarker of VK status for tissues that use MGP.

In addition to MGP, other VKD proteins, including growth-arrest specific gene 6, transforming growth factor β-induced protein ig-h3, periostin, Gla-rich protein and osteocalcin are present in cartilage and bone (12–15) so vitamin K may have multiple roles in joint health. To clarify what joint tissues VK is relevant to in OA, we investigated the cross-sectional and longitudinal association between plasma vitamin K, (dp)ucMGP and structural features of knee OA measured using MRI in older community-dwelling adults. We hypothesized that lower plasma vitamin K and higher (dp)ucMGP (reflective of lower vitamin K status) would be associated with higher prevalence and progression of knee OA features reported to be characterized by calcium deposition – namely articular cartilage and meniscus.

## METHODS

Participants were drawn from the Healthy, Aging, and Body Composition study (Health ABC), an ongoing prospective longitudinal cohort study designed to examine age-related changes in physical function and body composition in older black and white men and women. Between 1997 and 1998, 3075 well-functioning older black and white adults (40% black, 50% female, aged 70–79) were recruited from field centers in Pittsburgh, PA and Memphis, TN. At the time of recruitment, all participants were free of disability in activities of daily living and reported no difficulty walking ¼ mile or up 10 steps (16). The knee OA sub-study was initiated at the year 2 clinic visit (1998–99) when 640 participants with qualifying knee pain and 505 randomly selected controls underwent bilateral knee magnetic resonance imaging (MRIs). Cases with qualifying knee pain were identified if they had “knee pain, aching, or stiffness on most days for at least 1 month” at some point over the previous year or if they reported moderate or worse knee pain during the previous month in association with at least one activity on the WOMAC knee pain scale (17). Follow-up MRIs were completed at the year 5 clinic visit (median follow-up = 37 months) on 581 participants. Because warfarin is a vitamin K antagonist, participants in the knee OA sub-study who reported taking warfarin at the year 2 or year 5 clinic visits were excluded (n=65).

All participants provided written informed consent and the institutional review boards at both study sites approved all protocols.

## Knee image acquisition and reading

Bilateral knee MRIs were obtained using a Sigma 1.5T MRI system with a standard unilateral, commercial circumferential knee coil at the year 2 clinic visit and again 3 years later, as described (18). Coronal, sagittal, and axial images were obtained. Coronal views were T2-weighted fast spin-echo (FSE) (TR 3,500 msec, TE 20/60 msec) with a slice thickness of 4 mm, a 0.5-mm interslice gap, 2 excitation, FOV 14 cm, and a matrix of 256 × 256 pixels. Sagittal views were T2-weighted FSE, including the entire synovial cavity with frequency-selective fat suppression (TR 4,127 msec, TE 20/60 msec), a 0.5-mm interslice gap, 2 excitation, and the same FOV and matrix. Axial views were T2-weighted FSE (TR 2,500 msec, TE 20/60 msec) with a 1-mm interslice gap, 1 excitation, FOV 12 cm, and a matrix of 256 × 256 pixels). All follow-up images were read as a pair with their corresponding baseline image in sequence and have been evaluated semi-quantitatively using the Whole Organ MRI Score (WORMS) (19) by trained evaluators who were blinded to participant clinical history, at the University of California San Francisco's Arthritis Research Group. The inter-rater agreement for WORMS assessment by the 3 Health ABC MRI readers was good to excellent (intraclass correlation coefficient (ICC) = 0.63–0.93; with ICC<0.40 indicating poor agreement, 0.41–0.75 indicating fair to good agreement, and ICC >0.75 indicating excellent agreement) (20).

Thresholds used to define abnormal MRI knee OA features are summarized in Supplemental Table 1 (21). Articular cartilage lesion severity was scored on a 0–6 scale. Grade 1 lesions (signal abnormality) do not reflect morphologic change, so they were grouped with grade 0 for analysis. Osteophyte size was scored on a 0–7 scale. Grade1 osteophytes are considered equivocal, so were grouped with grade 0. Bone marrow lesions and subchondral cysts were each scored on a 0–3 scale for size, and subchondral bone attrition was scored on a 0–3 scale for severity. Synovitis/effusion was scored as grade 0 (normal), grade 1 (<33% of the maximum potential distension), grade 2 (33–66%), or grade 3 (≥66%) Meniscus damage was scored as grade 0 (intact), grade 1 (minor radial or parrot beak tear), grade 2 (nondisplaced tear or prior surgical repair), grade 3 (displaced tear, partial maceration, or partial resection), or grade 4 (complete maceration and destruction or complete resection) (19). At baseline each subregion was categorized as normal (grade 0) versus having an abnormality (lesion present) for each feature. Progression of each feature was defined as a score increase of at least 1 in any subregion. For articular cartilage damage and osteophyte progression, increases from grade 0 to 1 were not considered progression since grade 1 for those features indicates abnormal signal but not necessarily morphological change.

## Vitamin K Status

Blood samples were taken at the Year 2 clinic visit (1998/99) after an overnight fast and stored at −70°C until time of an alysis. Desphospho-ucMGP was measured from the same plasma samples using a sandwich ELISA, which uses 2 monoclonal antibodies directed against the nonphosphorylated amino acid sequence 3–15 and the noncarboxylated amino acid sequence 35–49 in human MGP. The reported intra- and inter-assay variability for this

assay were 5.6 and 9.9%, respectively (24). Plasma phylloquinone (vitamin K1, PK) was measured from stored samples using reversed-phase HPLC with post-column, solid phase chemical reduction of VK to its hydroquinone, followed by fluorometric detection at the Vitamin K Laboratory at the USDA Human Nutrition Research Center on Aging (HNRCA) at Tufts University (22). This laboratory currently participates in the international vitamin K external quality assurance scheme, KEQAS (23). The limit of detection for circulating PK with this assay using this sample volume available was< 0.2 nmol/L (22).

## Covariates

Body weight and height were measured using a standard balance-beam scale and Harpenden stadiometer (Holtain Ltd., Crosswell, UK), and body mass index (BMI) was calculated as weight (kg)/(height $(m)^2$). Interleukin-6 (IL-6) was measured in fasting serum in duplicate by ELISA (Quantikine HS; R & D Systems, Inc., Minneapolis, MN). Dietary intake over the previous year was estimated using an interviewer-administered 108-item Food Frequency Questionnaire (FFQ) developed specifically for Health ABC. The FFQ food list was derived using 24-hour recall data obtained from the National Health and Nutrition Examination Survey (NHANES) III for non-Hispanic black and white adults aged 65 and older residing in the Northeast or South. The Health ABC FFQ was analyzed for micronutrient and macronutrient content using the Block Dietary Data System (25). Physical activity was based on the reported time spent in walking for exercise or in other walking (eg, for transportation) over the previous week (25). The season during which the blood sample was obtained was included to account for seasonal effects on vitamin K. At the baseline clinic visit, demographic and lifestyle characteristics, including age, sex, race, education, smoking status and medication use were ascertained using an interviewer-administered questionnaire. Triglycerides were measured in fasting serum on a commercially available analyzer (Vitros 950; Johnson & Johnson, Rochester, NY). Clinic site was either Pittsburgh PA or Memphis TN.

## Statistical analyses

Given the novelty of plasma (dp)ucMGP and the lack of established normal ranges, categories were based on distribution quartiles. Plasma PK was categorized as <0.2 nmol/L (below the detectable limit), 0.2– <1.0 nmol/L or ≥1.0 nmol/L, because 1.0 nmol/L is the concentration that is generally achieved when adequate intakes (90 μg/day for women, 120 μg/day for men) are met (26;27). In descriptive analyses, differences in demographic, lifestyle and clinical characteristics were examined across categories of PK and (dp)ucMGP using general linear models for continuous outcomes or the Chi-square tests for categorical outcomes. Using the knee as the unit of analysis, the odds ratios (and 95% confidence intervals) [OR(95%CI)] for having abnormal knee OA features (based on thresholds defined in Table 1) according to categories of PK and (dp)ucMGP were calculated using the marginal model with generalized estimating equations (GEEs) to account for between-knee correlation within subject. Covariates included age, sex, BMI, race, smoking status, season of blood draw, education, and clinic site. We also adjusted for triglycerides and statin use because vitamin K is transported on triglyceride-rich lipoproteins (28) and may be affected by lipid lowering medications. Additionally, vitamin K status can reflect generally healthy lifestyles (29), so vegetable intake (an indicator of a healthy diet) and physical activity were

included as covariates. Since vitamin K has been shown to reduce production of pro-inflammatory cytokines (30), and inflammation has been implicated in OA (1), we included IL-6 as a covariate as well. Longitudinal models were additionally adjusted for follow-up time. Knees with the highest possible score at baseline for each feature were removed from longitudinal models to prevent a ceiling effect. We checked for non-linear associations between vitamin K status and each knee OA feature cross-sectionally and longitudinally by entering quadratic terms of the continuous exposures into each model (ie. plasma $PK^2$ or (dp)ucMGP$^2$). We also examined for effect modification by sex and race by entering product terms (ie. sex*PK category, sex*(dp)ucMGP quartile) into each model. Quadratic and interaction terms were considered significant at $p<0.05$. In sensitivity analyses we excluded participants without knee pain from cross-sectional and longitudinal analyses and excluded participants without prevalent features at baseline from longitudinal analyses. All analyses were carried out using SAS v 9.3 (SAS Inc., Cary, NC).

## RESULTS

After all exclusions, seven hundred ninety one (791) Health ABC knee OA study participants not taking warfarin had usable baseline knee MRIs. Measures of vitamin K status and pertinent covariates were analyzed cross-sectionally. Compared to participants excluded due to missing data (n=166), the included participants had a lower BMI (27.7±4.8 compared to 28.5±5.5 kg/m2, p=0.02), were more likely to be female (62% compared to 53%, p=0.01) and have graduated college (41% compared to 34%, p<0.01). Relatively more participants who reported knee pain were in the group excluded due to missing data as well (29% compared to 18%, p=0.01). Of the 791 participants with complete MRI and covariate data at baseline, 523 also had follow-up MRIs so were also analyzed longitudinally. Participants missing follow-up MRIs more likely to be male (59% compared to 47%, p<0.01), to be current smokers (11% compared to 8%, p=0.03), to not have completed education beyond high school (63% compared to 58%, p=0.01) and had higher IL6 levels (median 2.5 pg/ml vs 2.3 pg/ml, p=0.02). The prevalence of qualifying knee pain was similar between the two groups (72% compared to 75%, p=0.12) and they did not differ in any other pertinent characteristics.

Baseline characteristics according to (dp)ucMGP quartile are shown in Table 1. Plasma (dp)ucMGP was positively correlated with triglycerides and IL-6. Participants with lower (dp)ucMGP were more likely to be black, from the Memphis study site, not take statins and not have graduated high school (Table 1). Plasma PK was inversely correlated with plasma (dp)ucMGP. As expected, PK was positively correlated with triglycerides because vitamin K is transported on triglyceride-rich lipoproteins (Table 2). Participants with lower baseline PK were more likely to be male, be from the Pittsburgh study site and smoke. Green leafy vegetables are the primary dietary source of PK so, as expected, PK was positively correlated with vegetable intake.

### Cross-sectional results

The ORs (and 95%CIs) for having knee OA features assessed using MRI at baseline according to plasma (dp)ucMGP and PK categories are shown in Figure 1. Participants in

the higher quartiles of plasma (dp)ucMGP (reflective of lower vitamin K status) were more likely to have osteophytes, bone marrow lesions, subarticular cysts, and meniscal damage at baseline (Figure 1). The associations between plasma (dp)ucMGP and bone marrow lesions, subarticular cysts, and meniscal damage appear to be nonlinear (plasma (dp)ucMGP$^2$ terms $p<0.01$, $p=0.05$, and $p=0.04$ respectively; Figure 1). These cross-sectional findings were not appreciably different when analyses were restricted to participants with qualifying knee pain (n=558). None of the cross-sectional associations between plasma (dp)ucMGP and knee OA features on MRI differed by sex (all sex p-for-interaction>0.29). However, we detected an interaction between plasma (dp)ucMGP and race with respect to articular cartilage damage (p-for-interaction=0.02), such that blacks in the higher (dp)ucMGP quartiles were more likely to have articular cartilage damage at baseline [OR(95%CI), compared to Q1: Q2=1.3(0.8–2.3); Q3= 2.4(1.2–5.1); Q4=2.3(1.1–5.0)]. Among whites, (dp)ucMGP was not associated with articular cartilage damage at baseline [OR(95%CI), compared to Q1: Q2=0.7(0.4–1.3); Q3=0.9(0.5–1.6); Q4=0.8(0.5–1.4)].

Except for higher odds of prevalent subarticular cysts among those with non-detectable plasma PK, PK was not significantly associated with presence of abnormal knee OA features at baseline. The associations between PK and knee OA features on MRI did not have a quadratic component (all plasma PK$^2$ $p > 0.30$). None of the cross-sectional associations between PK and knee OA differed by sex (all sex p-for-interaction>0.09). We detected interaction between PK and race with respect to meniscal damage (p-for-interaction=0.01), such that whites with non-detectable plasma PK were more likely to have meniscal damage at baseline [OR(95%CI), compared to ≥1.0 nmol/L: non-detectable=2.6(1.3–5.9); detectable to < 1.0 nmol/L=1.0(0.7–1.5)]. PK was not associated with meniscal damage among blacks at baseline [OR(95%CI), compared to ≥1.0 nmol/L: non-detectable=0.7(0.3–1.5); detectable to < 1.0 nmol/L= 0.9(0.6–1.4)].

## Longitudinal results

Participants with non-detectable plasma PK at baseline were more likely to have worsening of articular cartilage damage and meniscal damage at three years of follow-up, (Figure 2). Compared to the lowest (dp)ucMGP quartile, participants in quartile 2 were more likely to have bone attrition progression. Otherwise knee OA progression did not appear to differ according to plasma (dp)ucMGP quartile (Figure 2). When analyses were restricted to participants with qualifying knee pain at baseline (n=558) the association between PK and worsening of articular cartilage damage became stronger: OR(95%CI)=2.0(1.1–3.7) among those with non-detectable PK. Otherwise, the cross-sectional and longitudinal results were not appreciably different in this sub-group. Results were also similar when participants without prevalent knee OA features at baseline were excluded. None of the associations between PK or (dp)ucMGP and knee OA feature progression had a quadratic component (all PK$^2$ $p>0.11$; all plasma (dp)ucMGP$^2$ $p>0.17$). Associations between PK and (dp)ucMGP and progression of knee OA features did not differ by sex or race (all interaction term p-values ≥0.06).

## DISCUSSION

We hypothesized higher plasma (dp)ucMGP (reflective of lower vitamin K status) would be associated with worse articular cartilage and meniscal damage because those joint tissues are susceptible to calcium deposition and mineralization (31). Higher (dp)ucMGP was associated with higher odds of meniscal damage cross-sectionally, but it was not associated with articular cartilage damage. Participants with higher (dp)ucMGP were also more likely to have osteophytes, bone marrow lesions, and subarticular cysts. Since OA is a disease of the whole joint, and MGP is also found in bone (10), these findings may indicate a role for MGP in subarticular bone turnover. However, our cross-sectional findings were not confirmed longitudinally ((dp)ucMGP was not associated with progression of any feature over 3 years), so reverse causation may be involved.

Older community-dwelling older adults with very low plasma PK (defined as below the assay limit of detection, <0.2 nmol/L) had a 1.7- and 2.6-fold higher odds of worsening cartilage damage and meniscal damage over 3 years. Our data also suggest participants with very low PK were more likely to have bone attrition, subarticular cyst, and osteophyte progression, but statistical significance was not reached [OR(95%CI)s: 1.9(0.9–3.6); 1.5(0.8–2.7); 1.5(0.8–2.8) respectively]. Because the circulating concentration of PK considered sufficient is not defined, we chose thresholds based on the assay limit of detection (<0.2 nmol/L) and the concentration generally achieved when recommended intakes are met (1.0 nmol/L). In Health ABC, only very low circulating PK was associated with worsening of any knee OA features. In the Multicenter Osteoarthritis Study (MOST), plasma PK <0.5 nmol/L was associated with a more than 1.5-fold higher risk for incident radiographic knee OA and a more than 2-fold increased risk for incident articular cartilage lesions over 30 months (5). In the Framingham Offspring Study, circulating PK <1.0 nmol/L was associated with a higher prevalence of hand and knee OA cross-sectionally (4). However, we did not find plasma PK to be associated with presence of any feature cross-sectionally. A similar inconsistency has been reported in other studies of vitamin K status and chronic disease. For example, serum PK was associated with progression but not prevalence of coronary artery calcium (a condition in which a role for vitamin K is suggested, based on similar mechanisms as OA) in community dwelling adults (32).

That very low plasma PK was associated with worsening articular cartilage and meniscal damage, but (dp)ucMGP was not associated with progression of OA features may be because VKs role in these pathologies is independent of its function as an enzymatic co-factor. The role of vitamin K as an enzyme cofactor is post-translational, hence one would predict that vitamin K determines carboxylation of the VKDP, but not expression. However, vitamin K was shown to regulate expression of articular cartilage matrix genes (including MGP) (33;34), so alternate roles for vitamin K in joint health are plausible. Misra and colleagues found MGP polymorphisms, but not circulating concentrations, were associated with hand OA in older adults (35), suggesting gene expression to be more relevant to OA than circulating MGP or its associated carboxylation status. Alternately, VKD proteins other than MGP are found in joint tissues (12–15), so carboxylation of these proteins by vitamin K may be more relevant to OA.

While associations between circulating vitamin K and cartilage damage/loss have been reported (5), our finding that low plasma PK was associated with meniscal damage is novel. Meniscal injury resulting from trauma is a strong risk factor for OA (36). But, meniscal degeneration also occurs in the absence of trauma, and becomes more prevalent with age (37). Age-related meniscal lesions are associated with knee OA, and are reported to occur even before cartilage damage is apparent (38;39). VKD proteins have not been studied in the meniscus so mechanisms underlying vitamin Ks association with meniscal degeneration are uncertain. Since the meniscus is fibro-cartilaginous tissue, it would follow that the same VKD proteins found in articular cartilage are also in the meniscus. However, the collagen content and non-collagenous proteins in meniscus are different from articular cartilage (40), so it is plausible VKD proteins in the two tissues likewise differ. As this has not been studied, our findings are hypothesis-generating.

Our study is strengthened by the longitudinal design and assessment of knee OA features using MRI, which provides a more detailed and comprehensive assessment of structural changes in the whole knee joint. To the best of our knowledge this is also the first assessment of vitamin K status and OA to use both plasma PK and (dp)ucMGP as indicators of status. However, the following limitations merit consideration. While circulating (dp)ucMGP reflects VK status, it is also dependent on the total amount of MGP in circulation such that the more total MGP synthesized, the more MGP there is to be carboxylated. This is an important distinction because both vitamin K status and total MGP (measured regardless of its carboxylation status) were independently identified as significant predictors of circulating (dp)ucMGP in older adults (11). Since total MGP has not been measured in Health ABC we were unable to adjust our models to express (dp)ucMGP as a ratio of total MGP, such as is the practice with other VKDP. Our hypothesis that vitamin K status would be associated with outcomes prone to calcification because of MGPs role as a mineralization inhibitor was supported by our findings that very low PK was associated with higher odds of meniscal and cartilage damage progression. However, calcium deposition was not directly scored on the MRIs.

We adjusted for vegetable intake and physical activity (time spent walking per week) to account for healthy lifestyles which can reflect vitamin K status (29), but residual confounding may persist. Health ABC participants were selected because they reported being well-functioning at 70–79 years old, and our findings may not be generalizable to dissimilar groups. Meniscal degeneration was likely more relevant to this cohort, given their age and which strengthened our ability to detect associations. Our results did not appear to differ in men and women, and the majority of our results were consistent between blacks and whites. We detected race interactions with respect to cartilage and meniscus damage cross-sectionally. Since we did not hypothesize associations would differ by race, type I error is a possible explanation. However, knee OA (41) and circulating PK (42) are reported to differ by race and ethnicity, and our data suggest (dp)ucMGP does likewise (Table 2), so these findings merit future investigation. There were some differences between participants who had follow-up MRIs and those missing follow-up MRIs, so the generalizability of our longitudinal findings may be limited. It is plausible those who did not return had more severe knee OA, so attrition bias merits consideration.

Consistent with the only other known longitudinal study of vitamin K and OA progression, (which found low plasma vitamin K was associated with more cartilage loss (5)), our results suggest vitamin K is implicated in progression of several distinct pathologies of OA affected joint tissues. Since plasma (dp)ucMGP was associated with knee OA features cross-sectionally but not longitudinally, future studies are needed elucidate mechanisms underlying vitamin Ks role in OA progression.

## Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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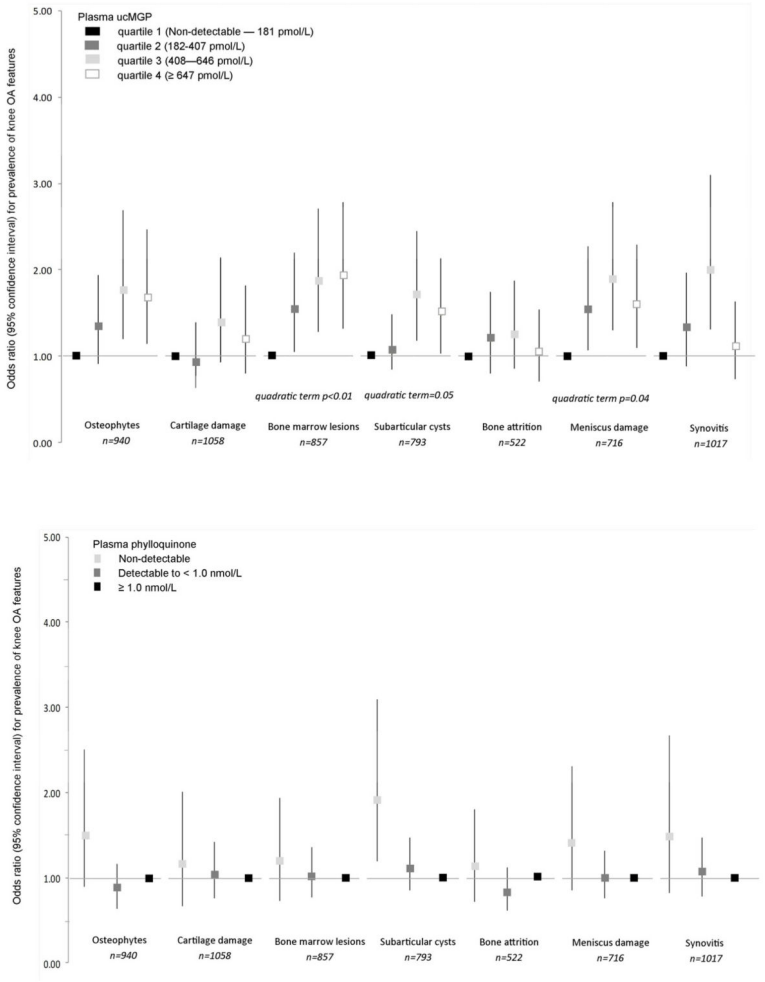


**Figure 1.**

Adjusted odds ratios with 95% confidence intervals* for prevalent knee OA features detected on MRI according to plasma (dp)ucMGP quartile (top) and phylloquinone category (bottom) and in 791 Health ABC participants

* adjusted for age, sex, BMI, race, smoking, vegetable intake, physical activity (time spent walking/week), triglycerides, statin use, interleukin-6, season of blood draw, education, and study site.

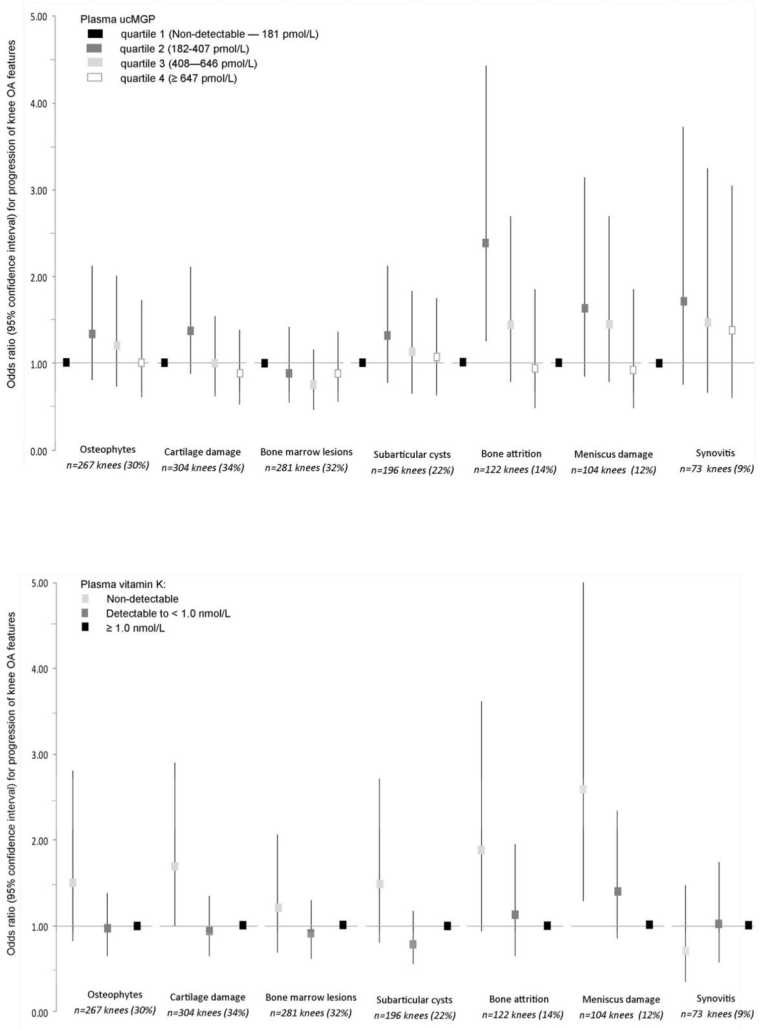


**Figure 2.**

Adjusted odds ratios with 95% confidence intervals* for worsening of knee OA features after 3 years detected on MRI according to plasma (dp)ucMGP quartile (top) and phylloquinone category (bottom) in 523 Health ABC participants.

* adjusted for age, sex, BMI, race, smoking, vegetable intake, physical activity (time spent walking/week), triglycerides, statin use, interleukin-6, season of blood draw, education, study site and follow-up time.

## Table 1

Baseline characteristics according to plasma (dp)ucMGP quartile of 791 Health ABC participants aged 70–79 y. Presented as median (interquartile range*), unless indicated otherwise

| (dp)ucMGP range | Q1: n=215 ND-181 pmol/L | Q2: n=193 182–407 pmol/L | Q3: n=192 408–646 pmol/L | Q4: n=191 ≥647 pmol/L |
|---|---|---|---|---|
| Age (yr) # | 74 (5) | 74 (4) | 74 (5) | 75 (4) |
| Female (n(%)) | 135 (63) | 118 (61) | 118 (61) | 122 (64) |
| Black (n(%)) # | 137 (64) | 92 (48) | 77 (40) | 60 (31) |
| BMI (kg/m$^2$) | 27.2 (6.5) | 26.7 (5.7) | 27.3 (5.2) | 27.9 (6.2) |
| Triglycerides (mg/dl) # | 112 (74) | 115 (56) | 122 (64) | 133 (77) |
| IL-6 (pmol/L) # | 2.2 (2.1) | 2.1 (2.2) | 2.5 (2.2) | 2.8 (2.9) |
| Phylloquinone (nmol/L) # | 0.9 (1.1) | 0.8 (0.8) | 0.8 (0.8) | 0.7 (0.7) |
| Vegetable intake (servings/day) | 3 (2) | 2 (2) | 3 (2) | 3 (2) |
| Physical activity (n(%)) min walking/week: | | | | |
| 0 | 96 (45) | 82 (42) | 79 (41) | 84 (44) |
| 1–150 | 79 (37) | 59 (31) | 55 (29) | 58 (30) |
| >150 | 40 (19) | 52 (27) | 58 (30) | 49 (27) |
| Smoker (n(%)) | | | | |
| current | 22 (10) | 19 (10) | 21 (11) | 9 (5) |
| former | 88 (41) | 80 (41) | 44 (85) | 85 (45) |
| Statin use (n(%)) # | 31 (7) | 56 (15) | 44 (12) | 44 (12) |
| Education (n(%)) # | | | | |
| < High school | 73 (34) | 42 (22) | 46 (24) | 35 (18) |
| High school graduate | 64 (30) | 60 (31) | 64 (33) | 82 (43) |
| College graduate | 78 (36) | 91 (47) | 82 (43) | 74 (39) |
| Season (n(%)) | | | | |
| Dec–Feb | 41 (19) | 51 (26) | 44 (23) | 48 (25) |
| Mar–May | 62 (29) | 51 (26) | 59 (31) | 55 (29) |
| Jun–Aug | 47 (22) | 38 (20) | 34 (18) | 36 (19) |
| Sept–Nov | 65 (30) | 53 (27) | 55 (29) | 52 (27) |
| Site (n(%) Pittsburgh) # | 90 (42) | 95 (49) | 113 (59) | 111 (58) |
| Qualifying knee pain (n(%)) | 150 (70) | 140 (73) | 131 (68) | 137 (72) |

* the difference between the 75$^{th}$ and 25$^{th}$ percentiles;

# $p \leq 0.05$ based on general linear models for continuous outcomes or the Chi-square tests for categorical outcomes

## Table 2

Baseline characteristics according to plasma vitamin K1 category of 791 Health ABC participants aged 70–79 y. Presented as median (interquartile range*), unless indicated otherwise

| | <0.2 nmol/L† (n=68) | 0.2–1.0 nmol/L (n=403) | ≥1.0 nmol/L (n=320) |
|---|---|---|---|
| Age (yr) | 75 (4) | 75 (5) | 74 (4) |
| Female (n(%)) # | 37 (54) | 240 (60) | 216 (68) |
| Black (n(%)) | 30 (44) | 186 (46) | 150 (47) |
| BMI (kg/m$^2$) | 27.0 (7.4) | 27.3 (5.6) | 27.7 (5.8) |
| Triglycerides (mg/dl) # | 102 (45) | 116 (58) | 131 (90) |
| IL-6 (pmol/L)# | 2.8 (2.9) | 2.4 (2.3) | 2.4 (2.3) |
| (dp)ucMGP (pmol/L) # | 311 (494) | 429 (481) | 344 (483) |
| Vegetable intake (servings/day) | 2 (2) | 2 (2) | 3 (2) |
| Physical activity (min walking/week) | | | |
| 0 | 29 (43) | 176 (44) | 136 (44) |
| 1–150 | 26 (38) | 123 (31) | 102 (32) |
| >150 | 13 (19) | 104 (26) | 82 (26) |
| Smoker (n(%))# | | | |
| current | 10 (5) | 41 (10) | 20 (6) |
| former | 33 (49) | 176 (46) | 129 (38) |
| Statin use (n(%)) | 14 (10) | 86 (11) | 75 (12) |
| Education (n(%)) | | | |
| < High school | 21 (31) | 97 (24) | 78 (24) |
| High school graduate | 19 (28) | 142 (43) | 109 (40) |
| College graduate | 28 (41) | 164 (41) | 133 (42) |
| Season (n(%))# | | | |
| Dec–Feb | 9 (13) | 109 (27) | 66 (21) |
| Mar–May | 5 (7) | 116 (29) | 106 (33) |
| Jun–Aug | 19 (27) | 61 (15) | 75 (48) |
| Sept–Nov | 35 (51) | 117 (28) | 73 (20) |
| Site (n(%) Pittsburgh) # | 44 (65) | 284 (60) | 138 (47) |
| Qualifying knee pain (n(%)) | 49 (72) | 284 (70) | 225 (70) |

* the difference between the 75$^{th}$ and 25$^{th}$ percentiles;

# p≤0.05 based on general linear models for continuous outcomes or the Chi-square tests for categorical outcomes;

† the lower detectable limit for this assay