Intake and serum concentrations of α-tocopherol in relation to fractures in elderly women and men: 2 cohort studies[1–3]

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ABSTRACT

Background: A reduction in the formation of free radicals and oxidative stress might reduce the rate of bone loss and muscle wasting.

Objective: The objective was to determine whether α-tocopherol intake or serum concentrations are associated with fracture risk in older women and men.

Design: Two cohort studies, the Swedish Mammography Cohort (SMC; $n = 61,433$ women) and the Uppsala Longitudinal Study of Adult Men (ULSAM; $n = 1138$ men), were used.

Results: During 19 y of follow-up, 14,738 women in the SMC experienced a first fracture at any site (3871 hip fractures). A higher hip fracture rate was observed with lower intakes of α-tocopherol. Compared with the highest quintile of intake, the lowest quintile had a multivariable-adjusted HR of 1.86 (95% CI: 1.67, 2.06). The HR of any fracture was 1.20 (95% CI: 1.14, 1.28). α-Tocopherol–containing supplement use was associated with a reduced rate of hip fracture (HR: 0.78; 95% CI: 0.65, 0.93) and any fracture (HR: 0.86; 95% CI: 0.78, 0.94). Compared with the highest quintile of α-tocopherol intake in ULSAM (follow-up: 12 y), lower intakes (quintiles 1–4) were associated with a higher rate of hip fracture (HR: 3.33; 95% CI: 1.43, 7.76) and any fracture (HR: 1.84; 95% CI: 1.18, 2.88). The HR for hip fracture in men for each 1-SD decrease in serum α-tocopherol was 1.58 (95% CI: 1.13, 2.22) and for any fracture was 1.23 (95% CI: 1.02, 1.48).

Conclusion: Low intakes and low serum concentrations of α-tocopherol are associated with an increased rate of fracture in elderly women and men. *Am J Clin Nutr* 2014;99:107–14.

INTRODUCTION

Osteoporotic fractures constitute a large and growing problem worldwide in both women and men, with a profound effect on quality of life (1) and mortality (2). Fracture risk is influenced by both genetic constitution and by environmental factors, and lifestyle factors (eg, diet and physical activity) become of greater importance with increasing age (3).

During the past decade, it has become evident that the increase in oxidative stress with aging is a fundamental pathogenetic mechanism of age-related bone loss (4) and also possibly sarcopenia (5, 6)—2 important determinants that contribute to the risk of fracture (7–9). Research suggests a progressive decrease in both bone and muscle mass by 1–2%/y after the age of 50 y (5).

A reduction in the formation of free radicals and oxidative stress might reduce the rate of bone loss and muscle wasting in the elderly (4, 5). Because α-tocopherol, the vitamin E com-

ponent with the highest antioxidant activity, is a potent scavenger of free radicals, it has been postulated that this form of vitamin E may favorably influence bone and muscle mass because of its antioxidant properties (4, 5, 10, 11).

However, human studies on the effect of α-tocopherol in relation to bone health are few. Importantly, there are no longitudinal data concerning the most relevant clinical endpoint of osteoporosis, fractures. We therefore examined the relation between α-tocopherol intake and fracture rate in elderly women within the Swedish Mammography Cohort (SMC)[4] (12) and validated the findings in the Uppsala Longitudinal Study of Adult Men (ULSAM) (13, 14)—2 community-based, longitudinal, and well-characterized cohorts.

SUBJECTS AND METHODS

Briefly, the SMC study started in 1987. All 90,303 women residing in 2 Swedish counties (Uppsala and Västmanland), born between 1914 and 1948, received a mailed invitation to a routine mammography screening. Enclosed with this invitation was a questionnaire covering diet (food-frequency questionnaire; FFQ) and lifestyle, which was completed by 74% of the women (12). In 1997, a subsequent expanded questionnaire was sent to those who were still living in the study area (response rate:

70%). The study sample with exclusions was described previously (12). In the current fracture study, 61,433 women with baseline data and 38,984 with data from 1997 were available for analysis. The studies were approved by the regional ethics committees at Karolinska Institutet, Stockholm, Sweden, and Uppsala University, Uppsala, Sweden, and all participants gave their informed consent.

Intakes of α-tocopherol and of other nutrients were estimated by multiplying the frequency of consumption of each food item by the nutrient content of age-specific portion sizes. The intakes were adjusted for total energy intake by using the residual method (15). α-Tocopherol intake estimated by the FFQ correlated with estimates from 14 repeated 24-h dietary-recall interviews evenly distributed over 1 y ($r = 0.57$) (16). Current use of vitamin E–containing supplements was reported in the second questionnaire.

The ULSAM cohort (13, 14) consists of 2322 men who were 50 y of age at enrollment in 1970–1974. The current analyses are based on the third examination cycle of the ULSAM cohort, when participants were ∼71 y of age (1991–1995). In all, 1138 men recorded their dietary intake during 7 consecutive days at the time of the third examination. Dietary habits were determined with an optically readable form of a 7-d dietary record that was based on a validated and previously used precoded menu book (17). The participants were given oral instructions by a dietitian on how to perform the dietary registration, and the amounts consumed were reported in household measurements or specified as portion sizes. The daily intake of α-tocopherol, energy, and other selected nutrients was calculated by using a database from the Swedish National Food Administration, which contains ∼1500 food items, drinks, and recipes. Serum α-tocopherol, adjusted for serum lipid concentrations (total serum cholesterol and triacylglycerols) (18), had been determined in a subgroup of 654 men, of whom 635 men had serum α-tocopherol also analyzed from samples collected at 50 y of age. HPLC was used for the analyses (19).

Fracture ascertainment

Fracture events in both cohorts were collated through linkage with the Swedish National Patient Registry and local outpatient registers within each county. The fracture collection in SMC began in 1987 and in ULSAM in 1991, and end of follow-up was 31 December 2010. Complete linkage with the register is rendered by the personal identity number provided to all Swedish residents. Any fracture event was defined as a hospital admission or an outpatient visit with an International Classification of Diseases 10th edition, diagnosis code of S12 through S92. Hip fracture cases were defined by codes S720 through S722. Incident fracture admissions were separated from readmissions (20), and only the first fracture event was used in the analysis.

Statistics

We estimated multivariable-adjusted HRs for risk of fracture by Cox proportional hazards regression in both cohorts. To minimize potential bias, the directed acyclic graph approach (21) was used to identify a suitable multivariable model. The multivariable models (adjusted for age, BMI, height, total energy and calcium intakes (all continuous), calcium and vitamin D sup-plementation (both dichotomous), educational level (≤9, 10–12, >12 y, or other), physical activity level [5 categories in SMC (12) and 4 in ULSAM (22)], smoking status (never, former, or current), estrogen replacement therapy (never, former, or current), and Charlson's comorbidity index (continuous) were similar in all analyses in both cohorts, although we additionally considered serum 25-hydroxyvitamin D and serum retinol concentrations (determined by HPLC, atmospheric pressure chemical ionization and mass spectrometry (APCI-MS), HPLC-APCI-MS, at Vitas, Oslo, Norway; www.vitas.no) at age 71 y, cognitive function, and FRAX scores (the WHO fracture prediction algorithm) (23) in ULSAM (23, 24). Other potential covariates (such as retinol, vitamin C, SFAs, MUFAs, PUFAs, alcohol, and protein intakes) in the multivariable models only marginally changed the relations and were therefore not included in the models. To better account for changes in the diet during follow-up and to better represent long-term dietary intake in the SMC, intakes of α-tocopherol and other nutrients estimated from the FFQs were treated as cumulative average intakes and the covariates as time-dependent variables (12). Covariates not assessed in the baseline FFQ of the SMC (eg, smoking habits and physical activity) were imputed by the Markov chain Monte Carlo multiple imputation method. Nonlinear trends of fracture risk were analyzed by quintiles of the exposure and by using a restricted cubic spline Cox regression model. We used 4 knots placed at percentiles 5, 35, 65, and 95. Finally, given that low vitamin E intake might be related to mortality, we both compared cumulative incidence curves with Kaplan-Meier failure curves (25) and estimated subhazard ratios of fracture according to the method of Fine and Gray (26) to address the potential competing risk problem from mortality. Even though these methods are not considered appropriate for etiologic analysis (27), which is the purpose of our analysis, competing risk analysis can provide additional valuable information for risk prediction (27).

Both low bone mineral density (BMD) and low muscle mass can be regarded as 2 component causes of fractures. In a supplementary mechanistic analysis, we therefore aimed to study the association between α-tocopherol intake and these components. Osteoporosis at the total hip, the femoral neck, or the spine diagnosed by dual-energy X-ray absorptiometry (DXA; Lunar Prodigy) was determined in a subcohort of SMC (SMC-Clinical; $n = 5022$) (12). Lean muscle mass was also determined by DXA, and skeletal muscle index (SMI) was calculated as the appendicular lean muscle mass (legs and arms) divided by height^2 (kg/m^2). Moreover, sarcopenia was defined as an SMI <5.45 (28). One to 3 months before the DXA scan, the participants responded to a third FFQ that included both dietary assessment and covariate information. The associations between α-tocopherol intake with BMD, lean muscle mass, osteoporosis, and sarcopenia were evaluated with linear and logistic regression. For these analyses, we used multivariable models as described above but additionally included serum 25-hydroxyvitamin D and retinol concentrations (determined by HPLC–atmospheric pressure chemical ionization–tandem mass spectrometry at Vitas, Oslo, Norway) as well as total fat mass assessed by DXA.

A flowchart describing SMC, SMC-Clinical, and ULSAM is displayed elsewhere (*see* Supplemental Figure 1 under "Supplemental data" in the online issue). Statistical analyses were performed by using Stata 11.2 (Stata Corp) and SAS version 9.3 (SAS Institute).

RESULTS

Fracture risk in women

Characteristics of the women in the SMC at baseline (1987–1990) are displayed in **Table 1**. With increasing energy-adjusted dietary α-tocopherol intake, only modest differences were observed in reported intakes of energy, calcium, and vitamin D. In addition, only small differences were noted in the number of comorbidities, educational level, physical activity, and smoking status between quintiles of dietary α-tocopherol intake.

During an average of 19 y of follow-up, 14,738 women experienced any type of first fracture (3871 of these were hip fractures). The multivariable-adjusted HRs for the risk of hip fracture (panel A) and any type of fracture (panel B) in the SMC in relation to energy-adjusted dietary intake of α-tocopherol are given in **Figure 1**. An exponential increase in hip fracture rate was discerned at intakes less than $\sim$5 mg/d, although the fracture rate was successively lowered also above this level without reaching a clear threshold. Compared with the highest quintile intake (median: 6.8 mg/d, **Table 2**), the lowest quintile intake (median: 4.3 mg/d) conferred a multivariable-adjusted HR of 1.86 (95% CI: 1.67, 2.06). The corresponding HR of any fracture was 1.20 (95% CI: 1.14, 1.28).

Moreover, current vitamin E–containing supplement use was reported by 10,801 of 38,984 women (28%) at the second questionnaire survey and was thereafter associated with a reduced rate of hip fracture (multivariable-adjusted HR: 0.78; 95% CI: 0.65, 0.93) and any type of fracture (HR: 0.86; 95% CI: 0.78, 0.94). The proportion of vitamin E supplement users was 30% in the highest baseline quintile of dietary α-tocopherol intake and 26% in the lowest quintile.

Fracture risk in men

Characteristics of the men in the ULSAM cohort at the third examination, by quintiles of α-tocopherol intake, are summarized in **Table 3**. Education, leisure physical activity, and energy, calcium, and vitamin D intakes tended to be higher in men with the highest quintile intake of α-tocopherol compared with lower intakes, whereas only small differences were seen in BMI and serum 25-hydroxyvitamin D concentrations. In addition, only 4% of the men reported use of any type of supplement. During an average follow-up of 12 y, 94 men had a hip fracture and 241 had any type of fracture. Low dietary intake was associated with a higher future fracture rate in men, but the association between dietary intake, α-tocopherol, and fracture rate was nonlinear (**Table 4**). Specifically, there was a tendency of higher rates of fracture observed in all of the first 4 quintiles compared with the highest quintile, although the estimates were not statistically significant for HRs of hip fracture in the 2 lowest quintiles. The lowest quintile intake of α-tocopherol, compared with the highest quintile, eg, was associated with a multivariable-adjusted HR for hip fracture of 2.23 (95% CI: 0.66, 7.54). The same comparison for any fracture conferred a statistically significant HR of 2.67 (95% CI: 1.33, 5.38). In comparison with the highest quintile intake (median: 8.1 mg/d), lower intake levels (quintiles 1–4) showed a multivariable-adjusted HR for hip fracture of 3.33; 95% CI: 1.43, 7.76) and an HR for any fracture of 1.84 (95% CI: 1.18, 2.88; data not shown in Table 4).

Characteristics of the men in ULSAM with serum α-tocopherol measurements ($n = 654$) are displayed in **Table 5**. During follow-up from age 71 y, we identified 51 men with an incident hip fracture and 139 with any type of fracture. Higher serum α-tocopherol was associated with a lower fracture rate, and the relation seemed to be linear for hip fracture ($P = 0.59$ for serum

TABLE 1
Some baseline characteristics of the women in the Swedish Mammography Cohort by quintiles of energy-adjusted dietary α-tocopherol intake

	Quintile 1 (<4.5 mg/d)	Quintile 2 (4.5–4.9 mg/d)	Quintile 3 (5.0–5.4 mg/d)	Quintile 4 (5.5–6.0 mg/d)	Quintile 5 (>6.0 mg/d)
No. of subjects	12,287	12,287	12,286	12,287	12,286
Age at entry (y)	53.1 ± 9.7[1]	53.1 ± 9.7	53.5 ± 9.7	54.1 ± 9.8	54.8 ± 9.7
BMI (kg/m^2)	24.6 ± 4.8	24.6 ± 4.0	24.7 ± 4.5	24.8 ± 4.0	25.2 ± 4.5
Height (cm)	159 ± 30	161 ± 25	161 ± 25	160 ± 26	160 ± 27
Dietary tocopherol intake (mg/d)	4.11 ± 0.37	4.77 ± 0.13	5.21 ± 0.13	5.72 ± 0.17	6.76 ± 0.70
Energy intake (kcal/d)	1560 ± 478	1614 ± 453	1604 ± 449	1577 ± 445	1563 ± 495
Calcium intake (mg/d)	972 ± 297	926 ± 248	903 ± 238	892 ± 235	857 ± 244
Vitamin D intake (mg/d)	4.02 ± 1.44	4.29 ± 1.23	4.41 ± 1.22	4.53 ± 1.27	4.82 ± 1.63
Calcium supplement use (%)[2]	14.2	16.2	17.4	17.6	16.5
Vitamin D supplement use (%)[2]	12.3	14.0	14.8	15.1	13.8
Vitamin E–containing supplement use (%)[2]	25.8	28.0	29.0	28.9	29.6
Hormone replacement therapy use (%)[2]	21.9	22.8	21.8	22.2	21.6
Low education, ≤9 y (%)	80.3	79.4	79.4	79.1	80.3
Cortisone use (%)[2]	0.1	0.2	0.1	0.2	0.2
Current smoker (%)	24.6	20.4	18.9	17.5	17.0
Former smoker (%)	30.7	29.6	29.1	29.8	30.1
Low physical activity (%)[2]	21.7	19.6	18.2	17.9	17.5
Nulliparous (%)	12.5	10.7	10.1	10.3	10.9
Previous fracture (%)	4.8	4.2	4.5	4.8	4.9
Living alone (%)	27.5	23.3	22.1	21.6	23.3

[1] Mean ± SD (all such values).
[2] Information available only in the 1997 questionnaire, and values are imputed from these data.
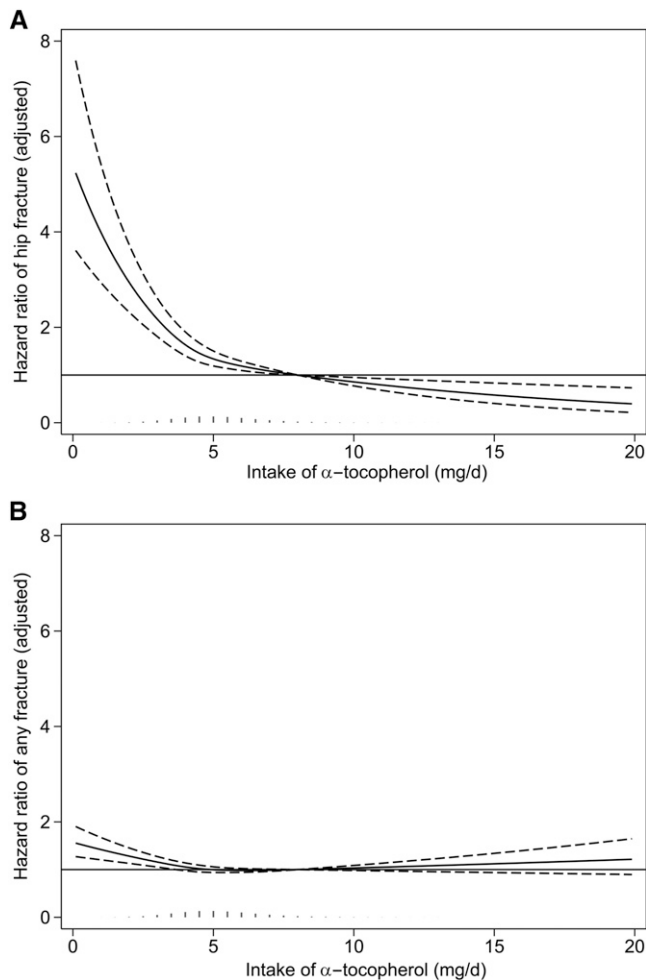
FIGURE 1. Multivariable-adjusted restricted cubic spline Cox's regression curve with 4 kn for the relation between dietary intake of α-tocopherol (the distribution is displayed by the rag plot above the x axis) and time to a first hip fracture (A) and any type of fracture (B) in women within the Swedish Mammography Cohort. The HR is indicated by the solid line and the 95% CI by the dashed lines. The reference was set at 8 mg/d, which is the recommended α-tocopherol intake for Swedish women. The HRs were adjusted for age, BMI, height, total energy intake, total calcium intake (all continuous), calcium (yes or no) and vitamin D (yes or no) supplementation, educational level (low, medium, high, or other), physical activity level (5 categories), smoking status (never, former, or current), and Charlson's comorbidity index (continuous).

α-tocopherol as a quadratic term) and for any fracture ($P = 0.70$). A 1-SD (0.30 mg/mmol) decrease in serum α-tocopherol concentration at age 71 y was associated with a multivariable-adjusted HR of 1.58 (95% CI: 1.13, 2.22) for hip fracture and of 1.23 (95% CI: 1.02, 1.48) for any fracture. Moreover, for each 1-SD reduction (0.34 mg/mmol) in serum α-tocopherol between ages 50 and 71 y, the HR of hip fracture was 1.71 (95% CI: 1.15, 2.53) and of any fracture was 1.27 (95% CI: 1.02, 1.58). These associations are also displayed as spline curves elsewhere (*see* Supplemental Figure 2 under "Supplemental data" in the online issue).

Mean ($\pm$SD) serum α-tocopherol concentrations in the highest quintile of dietary intake of α-tocopherol averaged 1.66 $\pm$ 0.29 mg/mmol compared with 1.60 $\pm$ 0.29 mg/mmol at lower intakes ($P = 0.04$). Only 28 men (2.5%) in the dietary survey reported use of vitamin E–containing supplements. They

also had, on average, 0.18-mg/mmol (95% CI: 0.06, 0.30) higher serum α-tocopherol concentrations ($P = 0.007$) than did non-users of vitamin E–containing supplements.

Competing risk

The cumulative incidence curves indicated that our results would not be compromised by competing risk from mortality (*see* Supplemental Figure 3 under "Supplemental data" in the online issue) in SMC and ULSAM (*see* Supplemental Figure 4 under "Supplemental data" in the online issue). Although we observed an excess mortality among women with a low α-tocopherol intake (*see* Supplemental Table 1a under "Supplemental data" in the online issue), the subhazard ratios of hip fracture were still elevated in these women (*see* Supplemental Table 1b under "Supplemental data" in the online issue).

α-Tocopherol intake, bone, and muscle

Characteristics of the subcohort SMC-Clinical are displayed elsewhere (*see* Supplemental Table 2 under "Supplemental data" in the online issue). Of these 5022 women with a mean age of 68 y, 1012 women had osteoporosis and 538 women were defined to have sarcopenia.

The intake of α-tocopherol intake was positively and independently associated with BMD and lean body mass. Thus, for each 3-mg decrease in α-tocopherol intake, the BMD of the proximal femur, after multivariable adjustment, decreased by 1.1% (95% CI: 0.3, 1.8; $P = 0.005$), the lumbar spine (L1-L4) by 0.8% (95% CI: -0.1, 1.8; $P = 0.09$), appendicular lean mass by 0.8% (95% CI: 0.2, 1.4; $P = 0.01$), and SMI by 0.6% (95% CI: 0.05, 1.18; $P = 0.03$). In addition, for every 3-mg decrease in α-tocopherol intake, the multivariable adjusted OR of osteoporosis at the lumbar spine was 1.20 (95% CI: 1.03, 1.41) and at the proximal femur was 1.46 (95% CI: 1.19, 1.78), and the OR of sarcopenia was 1.28 (95% CI: 1.04, 1.57). All P values of a quadratic term of α-tocopherol intake in the models were >0.05, which indicated linear associations.

DISCUSSION

In 2 independent cohorts we observed a higher rate of fracture in the elderly that was associated with low intakes of α-tocopherol. There was also a higher risk of both osteoporosis and sarcopenia with low intakes of α-tocopherol. Furthermore, the validity of these findings was corroborated by the relation between low serum α-tocopherol concentrations and a higher rate of fracture in men.

No previous prospective studies have assessed the relation between intakes or serum concentrations of α-tocopherol with future fracture risk. In previous cross-sectional studies, hip fracture patients had low vitamin E concentrations compared with control subjects at the time of the fracture event (29), and higher post-operative vitamin E concentrations were associated with lower concentrations of inflammatory markers (30). High serum concentrations of vitamin E were related to better physical function after the hip fracture (31). In animal models, supplementation with α-tocopherol improved fracture healing (10, 32–34). Interestingly, no association was found between BMD and vitamin E intake or serum vitamin E concentrations in the Women's Health Initiative Observational Study and Clinical Trial (35). However, it must be

TABLE 2
HRs for hip fracture and for any type of fracture derived from Cox proportional hazards models in relation to quintiles of dietary α-tocopherol intake in the Swedish Mammography Cohort[1]

	Quintile 1 (<4.5 mg/d)	Quintile 2 (4.5–4.9 mg/d)	Quintile 3 (5.0–5.4 mg/d)	Quintile 4 (5.5–6.0 mg/d)	Quintile 5 (>6.0 mg/d)
Hip fracture					
No. of fractures	591	574	692	906	1032
Duration of follow-up (person-years)	165,126	182,446	214,045	268,977	302,538
Crude rate (fractures/1000 person-years)	3.6 (3.3, 3.9)	3.2 (2.9, 3.4)	3.2 (3.0, 3.5)	3.4 (3.2, 3.6)	3.4 (3.2, 3.6)
Age-adjusted HR	2.03 (1.83, 2.25)	1.41 (1.28, 1.57)	1.19 (1.08, 1.31)	1.06 (0.97, 1.16)	1.00 (Reference)
Multivariable-adjusted HR[2]	1.86 (1.67, 2.06)	1.34 (1.21, 1.49)	1.17 (1.06, 1.29)	1.04 (0.98, 1.17)	1.00 (Reference)
Any fracture					
No. of fractures	1841	2011	2637	3617	4265
Duration of follow-up (person-years)	156,401	172,349	200,574	249,455	278,600
Crude rate (fractures/1000 person-years)	11.8 (11.2, 12.3)	11.7 (11.2, 12.2)	13.2 (12.7, 13.7)	14.5 (14.0, 15.0)	15.3 (14.9, 15.8)
Age-adjusted HR	1.27 (1.20, 1.34)	1.09 (1.04, 1.15)	1.05 (1.00, 1.10)	1.01 (0.97, 1.06)	1.00 (Reference)
Multivariable-adjusted HR[2]	1.20 (1.14, 1.28)	1.06 (1.01, 1.12)	1.03 (0.98, 1.08)	1.01 (0.96, 1.05)	1.00 (Reference)

[1] Values in parentheses are 95% CIs.
[2] The multivariable-adjusted HRs were adjusted for age, BMI, height, total energy intake, total calcium intake (all continuous), calcium (yes or no) and vitamin D (yes or no) supplementation, estrogen replacement therapy (never, former, or current), educational level (low, medium, high, or other), physical activity level (5 categories), smoking status (never, former, or current), and Charlson's comorbidity index (continuous).

emphasized that the participants had a total vitamin E intake averaging 29 mg/d (35), ie, substantially higher than that of the participants in the current investigation (mean: 5 mg/d). It must also be underscored that the rates of fracture in our studies were similar at intakes >10 mg/d, ie, no apparent further decrease in fracture rate was noted above this amount. Moreover, our group previously presented data indicating a negative influence of high oxidative stress and a beneficial effect of high vitamin E concentrations on bone within our Scandinavian setting (36–38).

Possible explanations and implications

Vitamin E has been proposed to have positive effects on both bone and muscle mass owing to its antioxidant properties (4–6,

10, 11), rendering a theoretical consequential lower fracture risk with a higher α-tocopherol intake. Our results support these experimental findings: higher intakes were associated with higher BMD, higher lean muscle mass, and lower fracture risk. In addition, recent randomized trials have indicated that vitamin E and C supplements have a positive effect on BMD and muscle mass in elderly women and men (39–41). Nevertheless, our results at first seem to contrast with experimental findings by Fujita et al (42), who recently found that a high intake of α-tocopherol was deleterious for bone health. However, the researchers fed 4-wk-old mice and rats a diet supplemented with 600 mg α-tocopherol/kg, which corresponds to a supplement that is almost 30 times higher than the amount recommended in rodents (43). Accumulating high doses of α-tocopherol given to

TABLE 3
Some baseline characteristics of the men in the Uppsala Longitudinal Study of Adult Men at age 71 y by quintiles of dietary α-tocopherol intake

	Quintile 1 (≤4.1 mg/d)	Quintile 2 (4.2–5.0 mg/d)	Quintile 3 (5.1–5.8 mg/d)	Quintile 4 (5.9–7.0 mg/d)	Quintile 5 (>7.0 mg/d)
No. of subjects	230	228	225	229	226
Age at entry (y)	71.0 ± 0.6[1]	71.0 ± 0.6	71.1 ± 0.7	71.0 ± 0.6	70.9 ± 0.7
BMI (kg/m²)	26.8 ± 3.5	26.6 ± 3.3	26.2 ± 3.2	26.1 ± 3.5	25.6 ± 3.3
Height (cm)	174 ± 6	175 ± 6	174 ± 6	175 ± 6	176 ± 6
α-Tocopherol intake (mg/d)	3.44 ± 0.51	4.57 ± 0.27	5.43 ± 0.24	6.39 ± 0.32	8.94 ± 2.90
Energy intake (kcal)	1269 ± 251	1514 ± 225	1731 ± 229	1940 ± 288	2284 ± 451
Calcium intake (mg/d)	734 ± 253	866 ± 267	960 ± 275	1055 ± 304	1221 ± 385
Vitamin D intake (mg/d)	3.87 ± 1.21	4.89 ± 1.33	5.58 ± 1.31	6.50 ± 1.67	7.96 ± 2.45
Serum 25-hydroxyvitamin D (nmol/L)	65.7 ± 18.4	69.8 ± 17.6	71.1 ± 20.7	70.3 ± 19.3	65.9 ± 17.1
Current smoker (%)	18.7	15.8	12.0	11.4	13.7
Former smoker (%)	50.9	51.8	55.6	45.4	43.8
Low education (%)	86.1	82.5	83.1	82.5	75.2
Cognitive impairment (%)	12.6	11.4	10.7	11.4	11.9
Secondary osteoporosis risk diseases (%)[2]	13.9	21.9	19.1	21.8	20.8
Previous fracture (%)	12.2	13.6	10.2	9.6	8.8
No Charlson comborbidity (%)	37.4	36.8	38.2	41.9	46.9
Low physical activity (%)	7.8	3.9	2.2	4.8	3.5
Living alone (%)	20.9	19.3	12.9	13.5	13.3

[1] Mean ± SD (all such values).
[2] Categorized as "yes" based on the presence of liver disease, type 1 diabetes mellitus, hypogonadism, malnutrition, or thyrotoxicosis.

TABLE 4
HRs for hip fracture and for any type of fracture derived from Cox proportional hazards models in relation to quintiles of dietary α-tocopherol intake in the Uppsala Longitudinal Study of Adult Men[1]

	Quintile 1 (≤4.1 mg/d)	Quintile 2 (4.2–5.0 mg/d)	Quintile 3 (5.1–5.8 mg/d)	Quintile 4 (5.9–7.0 mg/d)	Quintile 5 (>7.0 mg/d)
Hip fracture					
No. of fractures	18	20	25	23	8
Duration of follow-up (person-years)	2792	2831	2856	2929	3043
Crude rate (fractures/1000 person-years)	6.4	7.1	8.8	7.9	2.6
Age-adjusted HR	2.58 (1.12, 5.94)	2.87 (1.27, 6.52)	3.49 (1.57, 7.73)	3.10 (1.39, 6.93)	1.00 (Reference)
Multivariable-adjusted HR[2]	2.23 (0.66, 7.54)	2.84 (0.99, 8.21)	3.45 (1.33, 8.90)	3.02 (1.27, 7.21)	1.00 (Reference)
Any fracture					
No. of fractures	50	48	54	51	38
Duration of follow-up (person-years)	2612	2694	2680	2722	2857
Crude rate (fractures/1000 person-years)	19.1	17.8	20.1	18.7	13.3
Age-adjusted HR	1.52 (1.00, 2.33)	1.41 (0.92, 2.17)	1.63 (1.07, 2.47)	1.40 (0.91, 2.15)	1.00 (Reference)
Multivariable-adjusted HR[2]	2.67 (1.33, 5.38)	2.18 (1.19, 4.00)	2.30 (1.35, 3.93)	1.79 (1.10, 2.92)	1.00 (Reference)

[1] Values in parentheses are 95% CIs.

[2] The multivariable-adjusted HRs were adjusted for age, BMI, height, total energy intake, total calcium intake (all continuous), calcium (yes or no) and vitamin D (yes or no) supplementation, educational level (low, medium, high, or other), marital status (married or cohabitee or other), cognitive function (continuous), physical activity level (low, medium, or high), smoking status (never, former, or current), FRAX score (the WHO fracture prediction algorithm), Charlson's comorbidity index, and total serum 25-hydroxyvitamin D concentration (all continuous).

young rodents could be toxic (44) and lead to loss of appetite, reduced weight gain, and depressed bone growth. High dietary levels of vitamin E may also negatively affect the utilization of vitamin D_3 and lead to reduced bone mass (45). In addition, as shown in a meta-analysis of randomized studies, although low to moderate doses of vitamin E supplements might modestly reduce all-cause mortality (46), very high doses of vitamin E supplements (average dose: 400 mg/d) (46) may increase mortality in humans and is therefore not recommended (46). On the other hand, a corresponding dose given to rodents had anabolic bone effects (10). The highest percentile dietary α-tocopherol intakes in the SMC were 10 and 13 mg/d in ULSAM. We were therefore not able to consider the association between high-dose intakes of α-tocopherol and fracture risk.

The consumption of α-tocopherol is low in Sweden and other Scandinavian countries (47). Only ~10% of the women in the SMC and 5% of the men in ULSAM had an intake higher than the Swedish recommended intakes (8 and 10 mg/d for women and men, respectively). The highest incidences of osteoporotic fractures worldwide are also found in Scandinavia (48)—an observation that cannot readily be explained by known lifestyle or genetic determinants, climate, or longevity.

Strengths and weaknesses of the study

The strengths of the SMC were the size of the cohort, the population-based design, the large number of fractures, and the repeated FFQs. Because of the individual personal identification number of all Swedish residents, in combination with nationwide health care registers, we were able to identify almost all fractures that occurred in the cohort. However, we recognize that our study had limitations that should be considered when interpreting the results. First, one limitation common to observational studies is that they preclude conclusions regarding causality. Whether an interventional increased intake of α-tocopherol corresponds to a lower risk of fractures remains to be established. Importantly, findings from observational studies indicating that increases in

vitamin E intake might reduce cardiovascular disease, diabetes mellitus, and cancer have not been confirmed in randomized trials with very high intakes of α-tocopherol (49–51). We did have the possibility to adjust our estimates for comorbidities,

TABLE 5
Some baseline characteristics of adult men at age 71 y with serum α-tocopherol measurements in the Uppsala Longitudinal Study

	Value
No. of subjects	654
Age at entry (y)	71.2 ± 0.4[1]
BMI (kg/m²)	26.3 ± 3.4
Serum α-tocopherol (mg/L)	11.6 ± 2.8
Lipid-corrected serum α-tocopherol (mg/mmol)	1.61 ± 0.30
Serum triglycerides (mmol/L)	1.47 ± 0.76
Serum cholesterol (mmol/L)	5.81 ± 0.99
Serum α-tocopherol at age 50 y (mg/L)[2]	13.2 ± 3.2
Lipid-corrected serum α-tocopherol at age 50 y (mg/mmol)[2]	1.53 ± 0.28
Serum triglycerides at age 50 y (mmol/L)[2]	1.83 ± 0.83
Serum cholesterol at age 50 y (mmol/L)[2]	6.81 ± 1.22
α-Tocopherol intake (mg/d)	5.62 ± 1.90
Energy intake (kcal)	1736 ± 452
Calcium intake (mg/d)	979 ± 344
Vitamin D intake (mg/d)	5.74 ± 2.12
Serum 25-hydoxyvitamin D (nmol/L)	68.6 ± 19.4
Current smoker (%)	13.3
Former smoker (%)	45.1
Low education (%)	84.6
Cognitive impairment (%)	15.4
Secondary osteoporosis risk diseases (%)[3]	18.4
Previous fracture (%)	12.4
No Charlson comborbidity (%)	38.8
Low physical activity (%)	4.0
Living alone (%)	15.3

[1] Mean ± SD (all such values).

[2] $n = 635$.

[3] Categorized as "yes" based on the presence of liver disease, type 1 diabetes mellitus, hypogonadism, malnutrition, or thyrotoxicosis.

lifestyle factors, other nutrients, and a low vitamin D status (serum 25-hydroxyvitamin D concentrations), which is a frailty indicator in both men (52) and women (53), but the possibility of residual confounding still remains. In particular, people who use dietary supplements have, on average, a healthier lifestyle and a lower risk factor profile for cardiovascular disease (54), which could additionally lower their fracture risk (55). We considered the potential competing risk problem from mortality by the method of Fine and Gray (26) and by cumulative incidence curves (25). The subhazard ratios were similar to the HRs from the ordinary Cox regression, which suggests no major effect of competing risk, although it must also be emphasized that subhazard ratios are not directly comparable with ordinary cause-specific HRs (56). The latter estimates are the preferred measure of association in etiologic analysis (27)—the major focus of our study. Dietary-assessment methods are subject to many limitations that affect both the precision and accuracy of the measurement. An FFQ is used to assess the habitual intake of diet in larger studies such as the SMC, and a recent review concluded that it is a valid method for assessing dietary vitamin E intake (57). Nevertheless, the dietary recording in ULSAM can be regarded as a more precise dietary method with less misclassification (57), which, in combination with the homogeneous age in this cohort, may explain the somewhat stronger association with fracture rate in ULSAM compared with the SMC. However, we cannot exclude that these differences may reflect sex differences in the underlying pathology of osteoporotic fractures. By using the ULSAM cohort, although with a lower number of fractures rendering less precise estimates, we were able to replicate the findings from the SMC, which was a further strength of our study. Moreover, we were able to establish an association between serum concentrations of α-tocopherol and future fracture risk.

In conclusion, our observational data indicate that vitamin E insufficiency is associated with higher fracture risk. Additional studies are warranted to confirm our novel discoveries and ultimately randomized clinical trials that evaluate the effect on fracture risk of low doses (daily recommended intake) of vitamin E.

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