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# Wintertime Vitamin D Supplementation Inhibits Seasonal Variation of Calcitropic Hormones and Maintains Bone Turnover in Healthy Men

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**ABSTRACT:** Vitamin D is suggested to have a role in the coupling of bone resorption and formation. Compared with women, men are believed to have more stable bone remodeling, and thus, are considered less susceptible to the seasonal variation of calcitropic hormones. We examined whether seasonal variation exists in calcitropic hormones, bone remodeling markers, and BMD in healthy men. Furthermore, we determined which vitamin D intake is required to prevent this variation. Subjects ($N = 48$) were healthy white men 21–49 yr of age from the Helsinki area with a mean habitual dietary intake of vitamin D of 6.6 ± 5.1 (SD) μg/d. This was a 6-mo double-blinded vitamin D intervention study, in which subjects were allocated to three groups of 20 μg (800 IU), 10 μg (400 IU), or placebo. Fasting blood samplings were collected six times for analyses of serum (S-)25(OH)D, iPTH, bone-specific alkaline phosphatase (BALP), and TRACP. Radial volumetric BMD (vBMD) was measured at the beginning and end of the study with pQCT. Wintertime variation was noted in S-25(OH)D, S-PTH, and S-TRACP ($p < 0.001$, $p = 0.012$, and $p < 0.05$, respectively) but not in S-BALP or vBMD in the placebo group. Supplementation inhibited the winter elevation of PTH ($p = 0.035$), decreased the S-BALP concentration ($p < 0.05$), but benefited cortical BMD ($p = 0.09$) only slightly. Healthy men are exposed to wintertime decrease in vitamin D status that impacts PTH concentration. Vitamin D supplementation improved vitamin D status and inhibited the winter elevation of PTH and also decreased BALP concentration. The ratio of TRACP to BALP shows the coupling of bone remodeling in a robust way. A stable ratio was observed among those retaining a stable PTH throughout the study. A daily intake of vitamin D in the range of 17.5–20 μg (700–800 IU) seems to be required to prevent winter seasonal increases in PTH and maintain stable bone turnover in young, healthy white men.
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**Key words: vitamin D, PTH, ratio of TRACP to bone-specific alkaline phosphatase, pQCT, seasonal variation**

## INTRODUCTION

Knowledge of vitamin D in supporting bone health is rapidly growing. One of the remaining questions is does this apply only to the female skeleton. Most vitamin D interventions are carried out in female subjects, with the primary objective being to prevent osteoporosis. Because of the lower prevalence of male osteoporosis, the effect of vitamin D on the male skeleton has received less attention. Although the pathology of male osteoporosis differs from that of female osteoporosis, secondary hyperparathyroidism shares common features in both sexes.

In cross-sectional studies, an association between serum 25-hydroxyvitamin D [S-25(OH)D] and BMD has been reported among both men and women in different age groups.[1] Optimal bone health seems to be achieved with a vitamin D status of 80–100 nM.[2] However, only a few randomized controlled interventions of vitamin D either alone or together with calcium therapy have shown a positive effect on calcitropic hormones in men,[3,4] prevented bone loss among elderly men,[5,6] or decreased the risk for fractures.[5,7] Other studies have observed no benefits.[8,9]

Short-term deprivation of vitamin D stores is seen in the northern hemisphere because of scarce UVB light during winter accompanied by low dietary intakes of vitamin D,[10–13] complicating bone mineral accumulation in adolescents[14] and accelerating bone turnover in healthy adults.[15] Over time, this may contribute to a reduction in bone mass.[16] However, bone turnover among men and young subjects is speculated to be affected less by season than among elderly individuals.[17] This implies that the role of vitamin D in bone metabolism differs among sexes, warranting further research.

The objectives of this study were to characterize the adequate vitamin D intake for healthy men to maintain a

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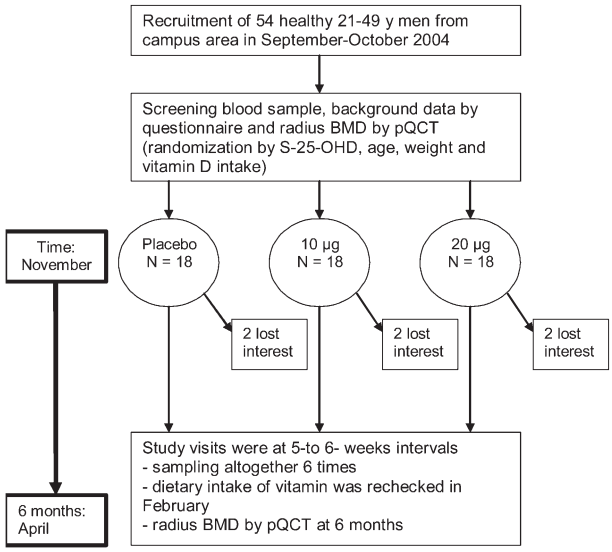


**FIG. 1.** The study flowchart.

stable postsummer vitamin D status throughout the winter and avoid a winter rise in serum PTH (S-PTH) concentration, and we investigated whether bone remodeling markers or the ratio of bone remodeling markers and radial volumetric BMD (vBMD) varied among these men during winter and whether these events could be prevented with vitamin D supplementation.

## MATERIALS AND METHODS

### *Subjects*

White men ($N = 54$) 21–49 yr of age were recruited from the Helsinki area (60° N) by an announcement in a campus area of the University of Helsinki, Helsinki, Finland. Power calculation based on S-25(OH)D (mean concentration, 68 ± 15 nM; expected change, 30 nM), assuming 90% power with $\alpha = 0.05$, concluded that a sample size of 12 was adequate. The study protocol was approved by the Ethics Committees of the hospital districts of Helsinki and Uusimaa. All subjects gave written informed consent in accordance with the Helsinki Declaration.

### *Protocol*

This was a randomized, double-blind, placebo-controlled, 6-mo vitamin D intervention study (Fig. 1). The 54 participants were allocated to three groups and assigned four tablets with morning meals daily consisting of 20 (800 IU) or 10 μg (400 IU) of vitamin $D_3$ or placebo. All tablets (Minisun) were provided by Verman (Järvenpää, Finland) and were similar in size and taste. The manufacturer confirmed that the actual vitamin D content of the pills was 7.5% higher than indicated in the labeling. Subjects were asked to take these tablets until trial closure and to return any unconsumed tablets during the last study visit to researchers for pill counting. In addition, the subjects were advised to record any days they had forgotten to take the pills in a follow-up diary. Compliance was confirmed with pill counts and the diary. Thus, the actual vitamin D intake from the supplements was 19.0 ± 4.5, 10.3 ± 0.4, and 0 μg, respectively. Of the 54 original participants, 2 subjects in each of the three study groups dropped out, leaving 16 subjects per group for a total of 48 subjects completing the 6-mo protocol. Traveling to sunny places or using tanning salons was not permitted during the study.

Randomization was performed to minimize the effect of variation in age, weight, initial S-25(OH)D status, and dietary intake of vitamin D on the three groups.

The trial lasted from the beginning of November until the end of April, for a total of 26 wk. Fasting blood samplings were collected at 5-to 6-wk intervals, altogether six times at the same time points between 7:30 and 9:30 a.m. Blood samples were processed within 3 h and centrifuged at 3000 rpm for 15 min, and serum was stored in aliquots at −70°C until analysis. Body weight, height, and distal and proximal radial BMD were measured by pQCT at enrollment and after 6 mo.

### *Laboratory measurements*

S-25(OH)D was measured from all fasting samples with an OCTEIA immunoenzymometric assay (IDS, Boldon, UK). The intra-assay CV was <2%. Interassay variation (7.9%) was avoided by measuring all samples from the same subject in the same series. Reproducibility was ensured by adhering to the Vitamin D External Quality Assessment Scheme, DEQAS. Standardized concentrations of S-25(OH)D were provided. Vitamin D status was defined as deficient at S-25(OH)D <25 nM, insufficient at 25 nM ≤ 25(OH)D < 80 nM, and sufficient at 25(OH)D ≥80 nM).[2]

S-PTH was measured from the first, third, and sixth repeated sampling with a commercial IEMA assay (IDS), with intra- and interassay CVs of 2.7% and 6.3%, respectively.

Serum bone-specific alkaline phosphatase (S-BALP) was assayed with an OCTEIA Octase BAP immunoenzymometric assay (IDS) to describe the bone formation in the first, third, and sixth repeated sampling. Intra- and interassay CVs were 2.9% and 5.0%, respectively.

The bone resorption marker, serum active isoform of 5b TRACP (S-TRACP), was determined with a Bone TRACP assay (SBA Sciences, Turku, Finland). Intra- and interassay CVs were 4.0% and 4.0%, respectively.

### *pQCT bone measurements*

pQCT was used to acquire peripheral BMD from the nondominant radius. Two 2.5-mm slices (voxel size, 0.5 mm), at the 4% and 66% sites, were measured proximally from the distal end of the radius (XCT-2000; Stratec, Pforzheim, Germany) and are referred to the distal and proximal radii in this text. A 30-mm planar scout view was used to locate a standard anatomical site for the radius reference line at the distal end. Length of the nondominant forearm was defined as the distance between the olecranon and the styloid process forming the basis for the location of the distal and proximal slices. After 6 mo, BMD measurements were performed with a repeated-measures program that allowed the starting point to be set according previous measurements.

TABLE 1. BASELINE CHARACTERISTICS

| | *Placebo* | *20 μg* | *10 μg* | *Significance* |
|---|---|---|---|---|
| *N* | 16 | 16 | 16 | |
| Age (yr) | 30.4 (7.2) | 28.0 (7.1) | 28.9 (6.8) | 0.605 |
| Height (cm) | 177.6 (6.7) | 181.1 (7.9) | 177.0 (8.0) | 0.227 |
| Weight (kg) | 78.7 (14.4) | 80.0 (16.1) | 78.4 (11.7) | 0.945 |
| Physical activity (min/d) | 43.3 (42.9) | 51.5 (41.7) | 50.9 (31.3) | 0.793 |
| Vitamin D intake (μg/d) | 6.6 (2.8) | 8.6 (6.3) | 7.6 (5.5) | 0.532 |
| Total vitamin D intake (μg/d) | 6.6 (2.8) | 27.8 (8.6) | 18.0 (6.0) | <0.001 |
| Calcium intake (mg/d) | 1250 (600) | 1510 (900) | 1260 (790) | 0.587[a] |
| S-25(OH)D (nM) | 64.7 (18.5) | 62.3 (13.6) | 60.3 (11.6) | 0.697 |
| S-PTH (pM) | 2.55 (0.62) | 2.73 (0.86) | 2.41 (0.77) | 0.476 |
| S-BALP (μg/liter) | 16.3 (4.6) | 21.9 (7.1) | 18.7 (5.2) | 0.054* |
| S-TRACP (U/liter) | 3.2 (0.7) | 3.6 (1.0) | 3.4 (0.7) | 0.389 |
| Distal radius | | | | |
| TB BMD (g/cm$^3$) | 368.0 (60.1) | 395.7 (48.8) | 379.7 (57.9) | 0.355 |
| TB CSA (mm$^2$) | 417.1 (75.8) | 385.4 (59.8) | 426.6 (78.2) | 0.225 |
| Trab BMD (g/cm$^3$) | 209.0 (42.7) | 222.2 (40.9) | 229.5 (42.9) | 0.365 |
| Proximal radius | | | | |
| Cort BMD (g/cm$^3$) | 1148.0 (17.6) | 1139.4 (35.3) | 1120.0 (30.7) | 0.021 |
| TB CSA (mm$^2$) | 99.4 (14.2) | 95.0 (14.9) | 105.2 (21.4) | 0.230 |

Values are mean (SD).
* Kruskal-Wallis test.

Data were analyzed using version 5.50 of the manufacturer's software package in which the outer contour of bone is defined with a threshold of 280 mg/cm$^3$.[18] The scans were analyzed using contour mode 2 (45%) and peel mode 1 to assess total (TB) and trabecular bone (Trab) parameters at the 4% site. At the 66% site, cortical bone (Cort) was detected with separation mode 1 and a threshold of 710 mg/cm$^3$.

Short-term precision (CV%) was determined with duplicate measurements of seven subjects. CVs for the BMD and the cross-section of area (CSA) in the TB, Cort, and Trab bone were 2.15, 1.99, 0.71, 0.88, and 1.32, 1.99, respectively. Phantom scans were executed daily to maintain quality assurance. The long-term CV% for the phantom BMD and CSA were 1.9, 1.1, 2.7, 0.79, and 0.50, 0.78 in the TB, Cort, and Trab, respectively.

### *Dietary assessment*

Dietary intake of vitamin D and calcium was assessed with validated food frequency questionnaires,[19] based on the Finnish national food database, Fineli. The subjects did not use other vitamin D–containing supplements than provided in the study. The major sources of dietary vitamin D were fish (32%), fortified milk products (25%), fortified spreads (18%), miscellaneous sources (18%), and eggs (7%).

### *Statistical analyses*

Statistical analyses were performed using SPSS version 12.0 for Windows (SPSS, Chicago, IL, USA). Pearson and Spearman correlations were used to assess the association between variables. Repeated-measures ANOVA was applied to evaluate the effect of supplementation in S-25(OH)D and S-PTH at different time points. The comparison of groups was performed with contrasts. Mean response was reported with area under curve (AUC), which was calculated with contrasts.

The change in BMD and bone remodeling markers was analyzed with analysis of covariance (ANCOVA), in which baseline value, calcium intake, physical activity, and corresponding change in the CSA were used as covariates. We found that ΔTB CSA correlated inversely with ΔTB BMD ($r = -0.888$, $p < 0.001$) at the distal site. ΔTB CSA confounds the TB BMD results at the 4% site, because the cortical layer thickness increases the farther from the distal site measured. Trab BMD is least confounded by ΔCSA.

Comparisons among several groups were done with ANOVA. Posthoc tests were carried out with least significant difference (LSD). If the variables were not normally distributed, the Kruskal-Wallis test was used.

Results are presented as mean ± SD, if not indicated otherwise. Results were considered significant when $p < 0.05$; $p$ values between 0.05 and 0.10 were considered trends.

The mean compliance was 94.5 ± 6.3%, and it did not differ among groups.

## RESULTS

Baseline characteristics are shown in Table 1. The median dietary intake from habitual diet was 6.6 ± 5.1 μg/d, which did not differ among groups. Total vitamin D intake includes habitual intake and compliance based intake from the study supplements. No change occurred in background characteristics (i.e., calcium intake and physical activity) during the study.

### *S-25(OH)D*

Figure 2A shows the response of S-25(OH)D to different doses of vitamin D. A significant difference was observed among groups with repeated-measures ANOVA ($p < 0.001$).

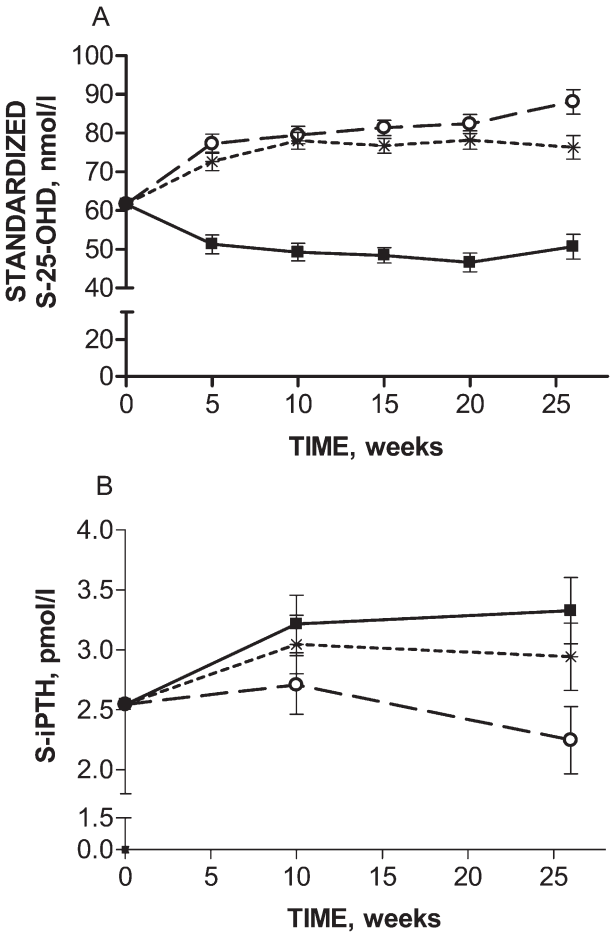


**FIG. 2.** (A) Response of S-25(OH)D concentration to vitamin D supplementation. Time points are at 5-wk intervals, starting at the beginning of November. Symbols (■) 0, (*) 10, and (○) 20 μg/d represent mean values at each time point, and the error bars = SE. A significant difference was present among groups with repeated measures ANOVA ($p < 0.001$). Baseline 25(OH)D was used as a covariate ($p < 0.001$). (B) Effect of supplementation on S-PTH concentration. Mean values are presented at the first, third, and sixth time points with error bars. A significant difference was present among groups with repeated-measures ANOVA ($p = 0.035$). Baseline S-PTH was used as a covariate ($p < 0.001$).

The mean response (AUC) was 27.0 ± 2.4 (SE) nM higher with 20 μg and 22.6 ± 3.4 (SE) nM higher with 10 μg than with the placebo ($p < 0.001$). The difference between the 10- and 20-μg groups was 4.5 ± 2.3 (SE) nM ($p = 0.064$).

From the first to the last time point, S-25(OH)D concentration increased 15.3 ± 14.4 and 27.8 ± 17.5 nM with a dose of 10 and 20 μg, respectively, whereas it decreased 12.5 ± 9.1 nM in the placebo group. ΔS-25(OH)D differed among study groups (ANOVA; $p < 0.001$). Mean dose-response [=ΔS-25(OH)D (nM)/dose(μg)] was 1.55 ± 1.24 nM/μg, and this did not differ between supplemented groups.

### *S-PTH*

The response of S-PTH to supplementation differed among the groups (repeated-measures ANOVA; $p = 0.035$; Fig. 2B). Although an increase occurred in the groups receiving placebo and 10 μg during the first 10 wk ($p = 0.014$ and $p = 0.043$, respectively), the overall response in S-PTH calculated from the first to last time point was null in the 10-μg group (repeated-measures ANOVA; $p = 0.140$), whereas a mean increase of 31% was observed in the placebo group ($p = 0.024$) and a mean decrease by 13.3% in the 20-μg group ($p = 0.012$).

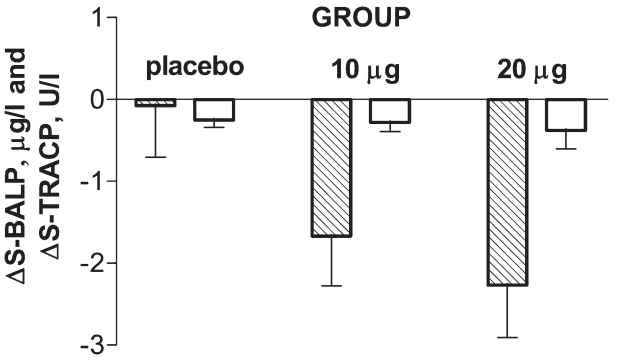


**FIG. 3.** Changes in bone remodeling markers during the study. Formation marker S-BALP (shaded bars) decreased more in the vitamin D–treated groups than in the placebo group (ANCOVA; $p < 0.05$). S-TRACP (white bars) acted similarly in all groups. Baseline values were used as covariates in the model.

### *Bone data*

No seasonal variation was observed in BMD in the placebo group ($N = 16$) during the 6-mo interval (data not shown).

*Distal radius:* Change in BMD of TB and Trab were analyzed with ANCOVA using calcium intake, physical activity, baseline BMD, and ΔTB CSA as covariates. No difference was observed in the ΔTB BMD among groups ($p = 0.397$) or in ΔTrab BMD ($p = 0.241$) in the distal radius.

*Proximal radius:* ΔCort BMD at the proximal site was analyzed with ANCOVA using calcium intake, physical activity, baseline BMD and ΔTB CSA as covariates. A trend for difference was seen in the ΔCort BMD among groups ($p = 0.090$) in the proximal radius. ΔCort BMD increased with increasing vitamin D supplementation.

### *Bone remodeling markers*

At baseline, the bone remodeling markers correlated positively with each other in each group similarly (for the combined groups: $r = 0.309$, $p = 0.031$). At 6 mo, the bone remodeling markers correlated positively in the 10- and 20-μg groups($r = 0.521$, $p = 0.038$ and $r = 0.502$, $p = 0.046$, respectively) but not in the placebo group.

S-BALP decreased in the groups receiving vitamin D supplementation but did not change in the placebo group; thus, ΔS-BALP differed among groups (ANCOVA; $p < 0.05$; Fig. 3). The difference between the placebo and 20-μg groups was 2.19 ± 0.90 (SE) μg/liter ($p = 0.02$) and for the placebo and 10-μg groups was 1.60 ± 0.88 (SE) μg/liter ($p = 0.07$). The resorption marker, S-TRACP, decreased by 7.8% in the placebo group ($p < 0.05$), and a similar pattern

TABLE 2. CHANGES IN S-25(OH)D AND RATIO OF BONE REMODELING MARKERS IN TERTILES OF ΔS-PTH

| | *Tertiles* | | | |
|---|---|---|---|---|
| | *I* | *II* | *III* | *Significance* |
| ΔS-PTH (pM) | −0.70 (0.36) | 0.06 (0.17) | 1.10 (0.67) | <0.001 |
| Range | −1.38; −0.18 | −0.15; 0.34 | 0.37; 2.5 | |
| ΔS-25(OH)D (nM) | 17.9 (14.4) | 9.6 (21.2) | −4.2 (22.4) | 0.020* |
| ΔTRACP/BALP | 0.01 (0.037) | −0.004 (0.030) | −0.027 (0.056) | 0.060 |
| Total vitamin D intake (μg) | 21.2 (10.7) | 16.9 (8.9) | 12.0 (7.0) | 0.027† |

Values are mean (SD).

* $p = 0.267$ for the comparison of I and II, $p = 0.064$ for II and III, and $p = 0.006$ for I and III.

† $p = 0.195$ for the comparison of I and II, $p = 0.150$ for II and III, and $p = 0.007$ for I and III.

was observed in the supplemented groups, with no differences being present among groups.

The ratio of remodeling markers has been suggested to be more informative than individual markers.[20] The ratio of TRACP to BALP shows the balance between markers or the coupling of markers in a robust way. The ratio of bone remodeling markers did not differ among groups significantly (repeated-measures ANOVA: $p = 0.130$), but it was found to fluctuate with serum PTH and 25(OH)D. In a regression model, ΔS-PTH explained 8.8% of the variation ($p = 0.041$) and the standardized β was −2.108, whereas ΔS-25(OH)D explained 6.3% of this variation ($p = 0.092$) and the standardized β was 0.251. The association between S-PTH response and bone remodeling markers was further exploited by dividing the response in S-PTH into tertiles (Table 2). These results indicate that the vitamin D intake among those maintaining both a stable PTH and ratio of TRACP/BALP throughout the winter was 17 μg/d (95% CI, 12.0–21.8 μg).

## DISCUSSION

A clear seasonal variation was observed in 25(OH)D and PTH, the former decreasing and the latter increasing throughout the winter. Interestingly, the bone resorption marker S-TRACP decreased 7.8% during the 6-mo trial, but neither S-BALP, a biomarker of bone formation, nor radial vBMD were affected. Bone turnover in men is considered more stable than in women.[21,22] In addition, male bone loss typically occurs because of decreased bone turnover, especially a reduced bone formation, rather than accelerated bone turnover.[22] In previous studies,[16,23,24] bone resorption has been found to be accelerated during winter because of an elevated PTH concentration. Although PTH increased 31% and subjects in the placebo group became vitamin D insufficient, the marker of bone resorption, TRACP, did not increase and, if anything, decreased. Physical activity[25] and increased calcium intake are shown to suppress bone resorption, but both of these were ruled out in this study. Hence, whereas S-TRACP is a very reliable marker of bone resorption in the follow-up of treatment and the prognosis of bone disease,[26] the ratio of bone markers may better illuminate the situation.

Seasonality in S-25(OH)D and PTH occurs because of decreased sun exposure, especially affecting Nordic countries, and when vitamin D fortification of foods is lacking.[10,11,15] This variation is proposed to interfere with bone metabolism and lead to lower BMD, which is one of the risk factors for osteoporosis.[16] Fracture rates are higher during wintertime,[27] probably not only because of icy and snowy weather that contributes to falls[28] but also to impaired vitamin D status, which affects both bone strength and muscle performance.[29] Seasonal variation in BMD has been shown in longitudinal follow-up[16,24] and in cross-sectional studies.[30,31] In this study, radial vBMD of healthy men showed no variation according to season. Recently, Meier et al.[16] reported bone loss of 1% in subjects of both sexes 33–78 yr of age during two successive winters as measured by DXA of the central skeleton. However, the seasonal bone loss in their study[16] was not statistically significant. Strengths of our study included pQCT, which allows changes in bone to be localized, and double-blinded study design, which differs from the protocol of Meier et al.[16] However, changes in the peripheral skeleton might vary from those in central sites.

Vitamin D supplementation improved S-25(OH)D concentration dose dependently. Mean dose-response was 1.55 ± 1.24 nM/μg, which is in line with previous studies.[32,33] The standardized concentrations resulting from doses of 10 and 20 μg were 78 and 88 nM, respectively, both reflecting optimal vitamin D status.[2] Stable PTH concentration was maintained with 10 μg, whereas PTH was suppressed from normal baseline value with 20 μg. Because PTH is the main regulator of bone remodeling, damping it might induce adynamic bone disease, which is typical among kidney patients,[21] but excess supplementation of calcium and vitamin D may also predispose to the disease. Adynamic bone disease is recognized as low bone turnover, which particularly affects trabecular bone and might increase fracture risk.[21]

Supplementation decreased BALP concentration, whereas it remained unchanged in the placebo group. BALP is generally thought to indicate the viability of osteoblasts, but it is also considered a marker of whole bone turnover.[34] Mineralization of the bone matrix parallels the maturation of osteoblasts, thus decreasing BALP.[21] Among vitamin D–deficient children, BALP concentration is typically elevated[35]; bone formation rate is high; and once describing vitamin D, the concentration of BALP decreases as mineralization occurs.[36] Another possible explanation is that the bone turnover rate was decelerated, as PTH became lower in vitamin D–supplemented groups.

The concentration of TRACP decreased in all groups in a similar way. The results of TRACP are discordant with earlier studies in which bone resorption increased during the winter.[15,16] However, the basal vitamin D intake among men was nearly 7 μg, and men in the placebo group did not become vitamin D deficient but only vitamin D insufficient. Thus, the drop in 25(OH)D was not high enough to induce bone resorption among men who typically have more stable bone turnover than women.[22,37] The ratio or index applying both remodeling markers has been speculated to be more informative than individual markers.[20] Our results support this, because the ratio of TRACP to BALP varied according to S-PTH and S-25(OH)D. The ratio did not change in the tertile retaining a stable S-PTH, in which the vitamin D intake was 17 μg/d (95% CI, 12.0–21.8), whereas it changed in the other S-PTH tertiles. Similarly, the stronger correlations between the remodeling markers observed in the vitamin D supplemented groups showed the effect of vitamin D on coupling.

Results concerning vBMD showed only a tendency toward an effect of vitamin D on Cort bone. Cort BMD was affected positively by vitamin D supplementation, and a dose-responsive trend was recognized. Previously, the study of Moyer-Mileur et al.[38] among preadolescent girls indicated that calcium and vitamin D therapy increased Trab vBMD in the distal tibia. To our knowledge, no other studies exist that have used pQCT for this purpose. In some reports,[39–41] a positive association between 25(OH)D and areal BMD among men was described, and a similar association was observed among young females.[19] We calculated retrospectively by power analysis at 90% of the power that it would have required a sample size of 37 for each group to detect a 2% change in Cort BMD. Our initial sample size calculation based on 25(OH)D, unfortunately, was inadequate for vBMD.

Recently, Vieth et al.[42] and other experts concluded that 600–800 IU (15–20 μg) of vitamin D is needed to maintain bone health. We conclude that a total daily intake of vitamin D in the range of 17.5–20 μg (700–800 IU) seems to be needed to prevent winter seasonal increases in PTH and maintain stable bone turnover in young, healthy white men. The current Nordic recommendation for adults (7.5 μg, 300 IU)[43] is inadequate in respect to bone health.

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