# Serum 25-hydroxyvitamin D concentrations in girls aged 4–8 y living in the southeastern United States[1–3]

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**ABSTRACT**

**Background:** Evidence suggests that adults and adolescents throughout the United States are at risk of poor vitamin D status. However, vitamin D concentrations in young American children have not been assessed.

**Objective:** The relations between serum 25-hydroxyvitamin D [25(OH)D] and bone were examined in prepubertal girls.

**Design:** In the present cross-sectional study, serum 25(OH)D concentration was assessed in 168 prepubertal girls aged 4–8 y living in the southeastern United States with the use of radioimmunoassay. Bone area, bone mineral content, and areal bone mineral density were measured from total body, lumbar spine, proximal femur, and forearm with dual-energy X-ray absorptiometry. Data were analyzed with analysis of variance, analysis of covariance, stepwise multiple regression, and partial correlations.

**Results:** The mean (±SD) serum 25(OH)D was 93.8 ± 28.1 nmol/L (range: 31.1–181.4 nmol/L). In a multiple regression analysis, race and season were the strongest predictors of vitamin D status. The black girls had lower mean 25(OH)D values than did the white girls ($P < 0.01$), and 25(OH)D values were significantly different in the total sample between the seasons ($P < 0.001$), ranging from 74.4 nmol/L during the winter months to 107 nmol/L during the summer. After adjustment for season, age, race, and body mass index, 25(OH)D values were negatively correlated with forearm bone mineral content ($r = -0.18$; $P = 0.02$).

**Conclusions:** Unlike prior reports of adults and adolescents living in the southeastern United States, vitamin D status was adequate in the children of the present study. 25(OH)D concentrations were not positively associated with higher bone mineral. *Am J Clin Nutr* 2006;83:75–81.

**KEY WORDS** Serum 25-hydroxyvitamin D, prepubertal girls, Georgia, race, season, bone mineral, body composition

## INTRODUCTION

Predominately indoor lifestyles, low dietary vitamin D intakes, and reduced capacity for skin synthesis of vitamin D have led to reports of higher prevalences of vitamin D insufficiency in older adults living in the United States. The most obvious public health implication is related to increased osteoporosis prevalence; however, the discovery of vitamin D receptors in a multitude of tissues has led to studies linking vitamin D insufficiency to other chronic, nonosseous diseases, such as cancer, multiple sclerosis, diabetes, and schizophrenia (1, 2). The long-term latency of these diseases complicates the prospect of identifying a definitive optimal range for vitamin D. Using functional indexes such as secondary hyperparathyroidism, calcium absorption, and glucose tolerance, many vitamin D experts estimate that a serum 25-hydroxyvitamin D [25(OH)D] concentration ≥80 nmol/L indicates vitamin D sufficiency in adults (3). By this criterion, over one-half of American adults have wintertime hypovitaminosis D, with older adults and persons with dark skin at the greatest risk (4). Reflecting these findings, the Dietary Guidelines for Americans (5) now recommend that older adults >70 y and persons with dark skin consume 1000 IU vitamin D/d, which is a substantial increase from the recommendations put forth in 1997 by the Food and Nutrition Board (6).

Because of the link between vitamin D and diseases that could develop throughout the human life span, considerable interest in the 25(OH)D status of younger populations has been generated (4, 7, 8). Sullivan at al (9) reported that 48% of a sample of 9–11-y-old girls ($n = 23$) living in Maine (mean latitude: 44°N) who were followed for 3 y had insufficient vitamin D concentrations [25(OH)D <50 nmol/L]. Data from the National Health and Nutrition Examination Survey (NHANES) III showed that 47% of a sample of 12–19-y-old girls who lived in sunny locations (mean latitude: 32°N) had wintertime vitamin D insufficiency [25(OH)D <62.5 nmol/L] (4). Moreover, reports that adolescents who have dark skin pigmentation or high body mass are at an increased risk of hypovitaminosis D raises concern for the vitamin D status of certain youth populations (4, 7, 8). Poor vitamin D status in youth could have consequences that compromises adult bone health (10); in fact, studies conducted in adolescent girls in Finland suggested that lower vitamin D status correlated with smaller gains in lumbar spine areal bone mineral

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density (aBMD) and lower measures of volumetric BMD (11–13).

Three studies conducted in Argentina, Tasmania, and northern Spain investigated vitamin D status in prepubertal populations and found vitamin D insufficiency [25(OH)D <50 nmol/L] in 10–80% of the children (14–16). To date, no studies have assessed vitamin D status in young children who reside in the United States. The purpose of the present study was to assess serum 25(OH)D concentrations in 4–8-y-old prepubertal girls living in northeast Georgia (latitude: 34°N), an area in the southeastern United States with sunlight available year-round. The study also aimed to identify predictors of vitamin D status and to investigate the association between vitamin D status and bone area, bone mineral content (BMC), and aBMD.

## SUBJECTS AND METHODS

### Study participants

A cross-sectional study was conducted to investigate the 25(OH)D status in black and white girls aged 4–8 y from the Athens, GA, area (latitude: 34°N) in the southeastern United States. The study used baseline data from the University of Georgia Childhood Bone Study, a prospective study that investigated the influence of artistic gymnastic training on bone (17). At baseline, all children had participated in limited or no organized physical activity (<12 wk). A physician assessed the pubertal stage of each participant by using the criteria described by Tanner (18) and determined that all participants were prepubertal (ie, stage I for breast and pubic hair development). Participant race-ethnicity was classified by parent identification of the participant as non–Hispanic white, non–Hispanic black, Hispanic, Asian, Asian-Indian, Native American, or any combination of the above. Because a primary focus of the present study was to examine racial differences, the participants from less represented ethnic groups ($n < 7$) were excluded, which left 120 non–Hispanic white girls and 48 non–Hispanic black girls from those who were originally recruited ($n = 203$). None of the participants had serious medical conditions or used medication known to change bone metabolism. The girls' parents indicated the total household income on a questionnaire by checking the appropriate salary range (<$10 000, $10 000–$19 999, $20 000–$29 999,...or >$100 000). Before participation, each participant and her guardian completed informed assent and consent forms, respectively. The Institutional Review Board for Human Subjects at the University of Georgia approved the study protocol.

### Testing protocol

Data collection was conducted between October 1997 and October 2000 at the University of Georgia. After the participants fasted overnight, blood samples were collected between 0730 and 1000. Within 1 wk of the initial blood draw, the participants returned to the laboratory for bone scans, anthropometric measures, and questionnaires that assessed demographic information, dietary intakes, and physical activity.

### Anthropometry

For anthropometric measurements, the participants wore light clothing and no shoes. Each participant's height and weight were measured to the nearest 0.1 cm and 0.25 kg with a wall-mounted stadiometer (Novel Products, Rockton, IL) and a calibrated double-beam balance scale (Fairbanks Scales, Kansas City, MO), respectively. Body mass index (BMI) values were calculated as weight (in kg)/height$^2$ (in m). To measure the BMI percentile for each child, BMI values were plotted on individual BMI-for-age charts (19).

### Serum 25(OH)D concentration measurement

After an overnight fast, blood samples were collected for analysis of serum 25(OH)D concentrations and were stored at −70°C until analyzed. Serum 25(OH)D concentrations were assayed by radioimmunoassay (DiaSorin Laboratories, Stillwater, MN) and run in duplicate. The inter- and intraassay coefficients of variation were 9.7–10.9% and 4.3–8.0%, respectively.

### Bone variables

Bone area (in cm$^2$), BMC (in g), and aBMD (in g/cm$^2$) of the total body, lumbar spine, nondominant total proximal femur, and nondominant forearm were measured by dual-energy X-ray absorptiometry (DXA; QDR-1000W; Hologic Inc, Waltham, MA). The lumbar spine was analyzed with the use of DXA low density spine software version 4.74 (Hologic Inc), whereas body fat composition was analyzed with the use of DXA pediatric whole body analysis software version 5.73 (Hologic Inc). Daily calibration was performed with a calcium hydroxyapatite and epoxy lumbar spine phantom embedded in a Lucite cube (Hologic c-caliber anthropometric spine phantom, model DPA-QDR-1). The laboratory CV was 0.27%, which was calculated from 365 scans over 5 y (20). In our laboratory, test-retest measurements with DXA in 6–10-y-old girls ($n = 10$) showed the following CVs for aBMD: total body, 1.2%; lumbar spine, 1.3%; total proximal femur, 1.6%; and forearm, 2.1%, and showed a CV of 2.0% for percent fat.

### Dietary intake

To assess energy (in kcal), calcium (in mg), and vitamin D (in $\mu$g) intake, the participants and their parents completed 3-d diet records, a method that was found to be valid and reliable for estimating energy and nutrient intakes in children (21–23). To ensure the accuracy of the records, a trained lab technician gave parents training with a 24-h recall questionnaire and provided food models and pictures of serving sizes. Diet records included time of eating, type and amount of food, and preparation method. The form specifically inquired about the consumption of calcium-fortified foods and nutritional supplements, and these values were subsequently integrated into the dietary intake data. A trained lab technician analyzed the diet records using Food Processor II (version 7.5; ESHA Research, Salem, OR). With a one-way random effects model, the intraclass correlations ($R$) for the average measures of 3 d of dietary intake in 6–10-y-old girls ($n = 10$) completed twice in a 2-wk period were calculated to be 0.47, 0.71, and 0.94 for energy, calcium, and vitamin D, respectively.

### Physical activity

Physical activity was assessed with accelerometers that measure the frequency and intensity of activity in counts/min [model 7164; Computer Science Applications, Fort Walton Beach, FL]. Strong correlations were found between accelerometry measures and energy expenditure in young children (24). The participants wore the accelerometers for 3 d—2 weekdays and 1 weekend day. The accelerometers were worn above the iliac crest of the

**TABLE 1**
Participant characteristics[1]

| Variable | White | Black | $P^2$ |
|---|---|---|---|
| Age (y) | 6.1 ± 1.6 [120][3] | 6.5 ± 1.6 [48] | 0.202 |
| Parent income level[4] | 4.8 ± 2.1 [118] | 3.3 ± 2.3 [47] | <0.001 |
| Anthropometric measurements | | | |
| BMI (kg/m$^2$) | 16.9 ± 2.3 [119] | 17.5 ± 3.1 [48] | 0.165 |
| Body fat (%) | 27.2 ± 7.8 [118] | 24.2 ± 9.5 [47] | 0.064 |
| 25(OH)D (nmol/L) | 99.2 ± 28.2 [120] | 80.4 ± 23.1 [48] | <0.001 |
| Bone area (cm$^2$) | | | |
| Total body | 1163 ± 220 [118] | 1267 ± 278 [47] | 0.025 |
| Lumbar spine | 28.9 ± 4.4 [118] | 31.5 ± 5.3 [47] | 0.004 |
| Proximal femur | 14.5 ± 3.8 [86] | 17.1 ± 3.8 [37] | 0.001 |
| Forearm | 5.8 ± 1.2 [119] | 6.9 ± 1.6 [47] | <0.001 |
| BMC (g) | | | |
| Total body | 760 ± 195 [118] | 910 ± 292 [47] | 0.002 |
| Lumbar spine | 15.6 ± 3.8 [118] | 18.7 ± 5.3 [47] | 0.001 |
| Proximal femur | 8.6 ± 3.1 [86] | 11.3 ± 3.5 [37] | <0.001 |
| Forearm | 2.1 ± 0.57 [119] | 2.7 ± 0.89 [47] | <0.001 |
| aBMD (g/cm$^2$) | | | |
| Total body | 0.647 ± 0.052 [118] | 0.705 ± 0.076 [47] | <0.001 |
| Lumbar spine | 0.534 ± 0.055 [118] | 0.583 ± 0.075 [47] | <0.001 |
| Proximal femur | 0.582 ± 0.077 [86] | 0.651 ± 0.076 [37] | <0.001 |
| Forearm | 0.349 ± 0.034 [119] | 0.380 ± 0.047 [47] | <0.001 |
| Dietary intake | | | |
| Vitamin D ($\mu$g) | 9.7 ± 5.7 [114] | 6.8 ± 5.5 [42] | 0.005 |
| Calcium (mg) | 920 ± 411 [114] | 726 ± 294 [42] | 0.002 |
| Multivitamin users (%) | 61 [118] | 32 [47] | 0.001 |
| Physical activity (counts/min) | 732 ± 233 [114] | 703 ± 237 [43] | 0.496 |

[1] 25(OH)D, 25-hydroxyvitamin D; BMC, bone mineral content, aBMD, areal bone mineral density.
[2] Tests of significance between the racial groups were based on 2-tailed independent *t* tests.
[3] $\bar{x}$ ± SD (all such values); *n* in brackets.
[4] Ordinal numbers represent range of income in US dollars (0 = <$10 000, 1 = $10 000–19 999 . . . 10 ≥ $100 000).

right hip, a placement that was considered the most effective for accelerometry assessment in children (25). The Computer Science Applications monitor recorded data for each participant during 1-min epochs, which generated 3-d averages of activity in counts/min.

### Statistical analyses

Data were analyzed with SAS version 8.2 (SAS Institute, Cary, NC). Descriptive statistics were calculated for all variables. A $P$ value $\leq 0.05$ was considered statistically significant. Two-tailed independent samples *t* tests were used to compare means between the black and white girls for all independent variables. A two-factor analysis of variance was used to quantify the effects of season and race on 25(OH)D and to measure whether these factors interact in this sample. To analyze differences in 25(OH)D concentrations between the races, an analysis of covariance was used, with control for age and season (which were identified as significant covariates by stepwise linear regression from a group of potential covariates that included race, age, season, BMI, household income, calcium intake, and physical activity). Partial Pearson's correlation coefficients between 25(OH)D and independent variables were computed, with control for necessary covariates. Nonconstant variance was observed in bone area, BMC, and aBMD variables; thus, analyses that involved these variables were weighted by the inverse of the square of that bone variable.

Because season of testing was not evenly distributed across the total sample, it was considered a potential confounding variable in all statistical models that adjusted for covariates. Means for most variables were significantly different between the black and white girls; thus, the participant characteristics and mean 25(OH)D concentrations were displayed separately for the black and white participants by season. In addition, partial Pearson's correlation coefficients are reported separately for the black and white participants when significantly different associations between the races were found.

## RESULTS

### Participant characteristics

The participant characteristics are presented in **Table 1**. The black and white girls did not differ significantly in age, BMI, body fat composition, or physical activity; however, significant differences were observed in 25(OH)D concentrations, parent incomes, dietary intakes, and bone area, BMC, and aBMD at all skeletal sites (Table 1).

The mean serum 25(OH)D concentration in the total sample was 93.8 nmol/L; the mean was 19% lower in the black girls than in the white girls (Table 1). None of the participants had a 25(OH)D concentration under the reference point for severe vitamin D deficiency (<27.5 nmol/L). With 50 nmol/L as the upper limit for vitamin D insufficiency or hypovitaminosis D, only a few participants that were tested in the winter ($n = 2$) or spring ($n = 2$), and none that were tested in the summer or fall, were considered vitamin D insufficient. A two-factor analysis of variance uncovered no strong evidence of an interaction between the

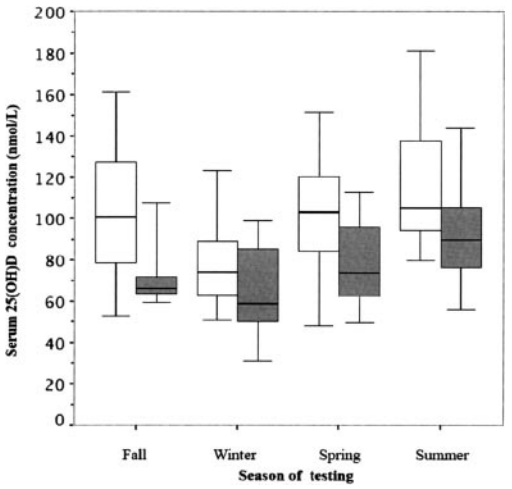


**FIGURE 1.** Box plot graph of serum 25-hydroxyvitamin D [25(OH)D] concentrations by race and season of testing. ■, black girls ($n = 5, 7, 22$, and 14 for fall, winter, spring, and summer, respectively); □, white girls ($n = 31$, 27, 40, and 22 for fall, winter, spring, and summer, respectively). Boxes represent values within the 25th–75th percentiles, and lines represent the range. Significant main effects of race and season were observed, but no significant interaction between race and season was observed. Means between seasons were significantly different for each race and marginally different when averaged over race ($P < 0.05$). Means between races were significantly different for spring, summer, and fall and marginally different when averaged over seasons (white girls > black girls; $P < 0.05$).

effects of race and season on the mean 25(OH)D concentration, but significant main effects of both factors were found ($P < 0.001$ for both race and season). The marginal mean 25(OH)D was significantly lower in the black girls than in the white girls, and predictable differences across seasons were observed; the mean serum 25(OH)D concentration of the participants that were tested in the winter was 32.1% lower than for those that were tested in the summer (**Figure 1**). The effect of season and race on 25(OH)D was reproduced in the multiple regression models in which race and season were identified as significant predictors of vitamin D status.

Mean vitamin D intake, calcium intake, and multivitamin use were higher in the white girls than in the black girls (Table 1). Mean vitamin D intake did not differ significantly across seasons, although it was higher in the winter than in the summer (10.6 compared with 8.1 $\mu$g/d; $P = 0.083$). Both races had mean dietary vitamin D intakes above the adequate intake level (5 $\mu$g/d), although the median intake in the black girls (median: 4.3 $\mu$g) was lower than the adequate intake. In the white girls, median daily intake of vitamin D was above the adequate intake (11.4 $\mu$g). Both the mean and median calcium intakes (median: 705 mg/d) in the black girls were slightly below the dietary reference intake for calcium (800 mg/d), whereas the mean and median calcium intakes (median: 863 mg/d) in the white girls were sufficient by this criterion.

### Partial correlations with 25(OH)D concentrations

#### *Age, household income, and dietary intake*

After adjustment for season and race, no significant associations were found in the total sample between 25(OH)D concentrations and household income, BMI percentile, body fat percentage, physical activity, or dietary vitamin D (**Table 2**). However, a significant association was found between 25(OH)D and dietary calcium. None of these partial associations was found to depend on race, but the association between 25(OH)D and age did differ significantly between the black and white girls, with a strong negative relation found for the black girls ($P = 0.005$) but not for the white girls. Although not statistically significant, positive relations were found in the total sample between 25(OH)D concentrations and both household income ($P = 0.054$) and dietary vitamin D intake ($P = 0.073$).

**TABLE 2**
Partial correlation coefficients ($r$) of serum 25-hydroxyvitamin D concentrations with multiple variables

| Variable | $r$ | $P$ |
|---|---|---|
| Age[1] | | |
| White | −0.023 | 0.810 |
| Black | −0.411 | 0.005 |
| Household income[2] | 0.152 | 0.054 |
| BMI percentile[2] | −0.010 | 0.899 |
| Body fat (%)[2] | −0.062 | 0.435 |
| Vitamin D intake ($\mu$g)[3] | 0.147 | 0.073 |
| Calcium intake (mg)[3] | 0.169 | 0.038 |
| Physical activity[2] | 0.055 | 0.501 |

[1] Partial correlation coefficients adjusted for season and computed separately by race because of a significant age-by-race interaction, $P = 0.04$.
[2] Adjusted for season and race.
[3] Adjusted for season, race, and energy intake.

#### *Bone variables*

The serum 25(OH)D concentration was negatively correlated with bone area, BMC, and aBMD at all sites (**Table 3**). However, after adjustment for season, race, age, and BMI, no significant associations remained except for forearm BMC (Table 3). This inverse association remained after additional adjustment for physical activity and calcium intake, although it was no longer statistically significant (Table 3).

## DISCUSSION

Vitamin D insufficiency would not be expected to be problematic for children living at a latitude of 34°N; however, little data are published to confirm this. The present study is the first to report serum 25(OH)D concentrations in young children living in the southeastern United States. Vitamin D insufficiency was not evident in these young girls; however, there were clear seasonal and racial differences in mean serum 25(OH)D concentrations. Although vitamin D is necessary for optimal calcium absorption and bone mineral accrual, no positive relations were observed between serum 25(OH)D concentrations and lumbar spine, proximal femur, or forearm BMC.

The findings of the present study contribute to existing evidence of latitudinal differences in the circulating 25(OH)D concentrations of children. Mean serum 25(OH)D values for these participants who lived at 34°N were higher than those of children who lived at higher latitudes in Spain (43°N) (16) and Tasmania (42°S) (15). Furthermore, only 2% of the participants in the present study had vitamin D insufficiency, whereas the prevalence was 12% (in the summer) and 80% (in the winter) in the children who lived in Spain (16) and 10% (in both the spring and winter) in the children from Tasmania (15) when using the present study's 50 nmol 25(OH)D/L cutoff. In NHANES III,

**TABLE 3**
Partial correlation coefficients (*r*) of serum 25-hydroxyvitamin D concentrations with bone area, bone mineral content (BMC), and areal bone mineral density (aBMD) in the total sample

| | Pearson correlations | | Partial Correlations *adjusted for season, race, age, and BMI* | | Partial Correlations *adjusted for season, race, age, BMI, calcium intake, and physical activity* | |
|---|---|---|---|---|---|---|
| Variable | *r* | *P* | *r* | *P* | *r* | *P* |
| Bone area ($cm^2$) | | | | | | |
| Total body | −0.204 | 0.009 | −0.003 | 0.969 | −0.025 | 0.773 |
| Lumbar spine | −0.226 | 0.004 | −0.088 | 0.271 | −0.093 | 0.277 |
| Forearm | −0.240 | 0.002 | −0.125 | 0.114 | −0.132 | 0.120 |
| Proximal femur | −0.251 | 0.005 | −0.007 | 0.943 | −0.006 | 0.949 |
| BMC (g) | | | | | | |
| Total body | −0.224 | 0.004 | −0.025 | 0.759 | −0.019 | 0.825 |
| Lumbar spine | −0.235 | 0.002 | −0.031 | 0.699 | −0.024 | 0.780 |
| Forearm | −0.263 | 0.001 | −0.182 | 0.021 | −0.154 | 0.069 |
| Proximal femur | −0.249 | 0.005 | 0.048 | 0.605 | 0.070 | 0.467 |
| aBMD ($g/cm^2$) | | | | | | |
| Total body | −0.213 | 0.006 | −0.021 | 0.793 | −0.012 | 0.886 |
| Lumbar spine | −0.207 | 0.008 | 0.005 | 0.951 | −0.001 | 0.987 |
| Forearm | −0.218 | 0.005 | −0.087 | 0.279 | −0.035 | 0.681 |
| Proximal femur | −0.170 | 0.061 | 0.129 | 0.166 | 0.135 | 0.159 |

white women aged 12–29 y who lived in the southern United States at latitudes ranging from 25–34.9°N had mean wintertime serum 25(OH)D values comparable to the white girls in the present study who live at a similar latitude (74.8 nmol/L compared with 76.7 nmol/L) (4). Serum concentrations of 25(OH)D from both the NHANES III survey and the present study were assessed by radioimmunoassay from the same manufacturer.

Despite the effect of latitude on circulating 25(OH)D concentrations, a recent review highlighted the fact that living at a low latitude does not entirely prevent poor vitamin D status (26). For example, in children who lived in Argentina (34°S), which is the latitudinal equivalent of our study locale, the wintertime 25(OH)D values ($\bar{x}$: 53 nmol/L) were 28% lower than the wintertime values of our population (14). Differences in vitamin D fortification in foods may account for the lower mean 25(OH)D values in Argentina. Black women in the NHANES III survey who lived in the southern United States at latitudes ranging from 25–34.9°N had mean wintertime serum 25(OH)D values that were lower than those of the black girls from the present study (42.3 nmol/L compared with 65.7 nmol/L). Furthermore, 70% of the black women in the NHANES survey had 25(OH)D concentrations <50 nmol/L compared with only 2% of the participants in the present study.

The relatively high mean serum 25(OH)D concentrations that were observed in the present study may be related to the young age of the participants. Previous studies that assessed vitamin D status in children noted that older age groups had a lower vitamin D status than did younger age groups (4, 27). An age-related decline in cutaneous synthesis of vitamin D may partly account for this, although the age when this decline occurs is unknown (28). The negative correlation between age and 25(OH)D in the black girls of the present study may not be related to impaired synthesis of vitamin D with age, but rather with an increase in time spent indoors as children advance in age (29, 30). Such a behavior change is supported by prospective studies that have reported an increase in sedentary activity with age, especially in black female adolescents (29, 30). If black children have the potential to develop hypovitaminosis D in adolescence and early adulthood, which is consistent from findings in 12–29-y-old black women (4), the recommendation by the Dietary Guidelines for Americans of 1000 IU vitamin D/d for persons with dark skin (5) becomes particularly important. Vitamin D intakes for the black participants in the present study fell substantially below this recommendation.

Despite the availability of year-round sunlight in the southeastern United States, mean 25(OH)D concentrations were 32% lower in the winter than in the summer, with season being the strongest predictor of vitamin D status. This finding is consistent with a recent study of adolescent girls living in the northeastern United States at a mean latitude of 44°N (9). The authors reported a 28% drop in 25(OH)D concentrations from September to March, which reflects summer and winter concentrations, respectively. After season, race had the strongest association with vitamin D status in our study. This observation is consistent with population studies reporting that persons with more melanin content had lower vitamin D status than did persons with less skin pigmentation (4, 7, 8, 31). Despite the observation that black girls had 21% lower mean 25(OH)D concentrations than did white girls, the black girls had higher BMC values at all skeletal sites.

No significant positive associations were found between 25(OH)D concentrations and total body, lumbar spine, or proximal femur BMC after adjustment for covariates. Lehtonen-Veromaa et al (32) found that vitamin D status was positively related to changes in aBMD accrual over 3 y, but only in more mature girls, ie, those who were in pubertal stages 3–4. The more advanced maturational period of these girls was likely important with respect to their bone mineral acquisition (the girls in our study were in pubertal stage 1). Unfortunately, the authors did not report site-specific changes in BMC, which is a more appropriate measure for studies of growing children and adolescents. Similar to our finding of a negative association between forearm BMC and serum 25(OH)D concentrations, Cheng et al (11) reported that participants with sufficient vitamin D status had significantly lower proximal femur BMC than did women with insufficient vitamin D

status. The fact that our participants were young, were in a relatively slower period of bone growth, had adequate calcium intakes, and had high serum 25(OH)D concentrations may partially account for the lack of positive associations.

One limitation of the present study was that we did not measure parathyroid hormone (PTH). Researchers in the vitamin D field agree that the concentration at which vitamin D suppresses PTH can be used as a functional index of sufficient vitamin D status (33). Unfortunately, evidence of the threshold for optimal vitamin D status is conflicting, especially in children (7). Previous studies have observed suppression of PTH at 25(OH)D concentrations between 30 and 50 nmol/L in children aged 7–10 y and between 40 and 60 nmol/L in adolescents (7, 13, 16, 34). If we extrapolate that 50 nmol/L indicates optimal PTH suppression in our study age group, only 2% of our sample would have been at risk of vitamin D insufficiency. It is important to note the limitation of comparing 25(OH)D data in the present study to other studies (35, 36). The methods that are used to assess serum 25(OH)D concentrations vary between laboratories and can yield different values compared with the radioimmunoassay (DiaSorin) that was used in the present study.

In the temperate climate in which the present study was conducted, children seem to have a year-round sufficiency of vitamin D, although mean 25(OH)D values still differed depending on the season of testing. Direct measurements of UV exposure may help measure the amount of UV exposure needed to ensure optimal vitamin D status in different population groups. The negative association between 25(OH)D and age in the black girls may imply a future risk of hypovitaminosis D, possibly during puberty, which is the most critical period for bone mineralization (37, 38). Prospective studies are needed to address the changes in vitamin D status in black adolescents and the effect on bone mineral accrual.

RDL, CMM, MGK, MAJ, EML, and EMS were responsible for the study concept and design. RDL, CMM, EML, and ARW were responsible for supervising the acquisition of the data. DBH was responsible for the serum 25(OH)D analysis. DBH conducted the statistical analyses. RDL, EML, EMS, MGK, and MAJ were responsible for the interpretation of the data and the drafting the manuscript. All authors contributed to the revision of the manuscript. None of the authors had any personal or financial conflicts of interest.

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