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ORIGINAL ARTICLE

Serum 25-hydroxyvitamin D concentration and physical function in adult men

Lisa Ceglia*,†, Gretchen R. Chiu‡, Susan S. Harris† and Andre B. Araujo‡

*Division of Endocrinology, Diabetes and Metabolism, Tufts Medical Center, †Bone Metabolism Laboratory, Jean Mayer United States Department of Agriculture Human Nutrition Research Center on Aging at Tufts University, Boston and ‡Department of Epidemiology, New England Research Institutes, Inc., Watertown, MA, USA

Summary

Objective Recent reports suggest that vitamin D status influences musculoskeletal health; yet, there are limited data in adult men. This study investigated whether serum 25-hydroxyvitamin D [25(OH)D] concentration was associated with lean body mass, muscle strength and physical performance in men.

Design Population-based, observational survey.

Participants 1219 black, Hispanic and white randomly selected men aged 30–79 years from the Boston Area Community Health/Bone Survey.

Measurements Lean body mass by dual-energy X-ray absorptiometry, hand grip strength, a composite physical function score (chair stand and walking speed), 25(OH)D, parathyroid hormone (PTH), testosterone, age, race, body mass index, socioeconomic status, education, smoking, arthritis, self-reported health, calcium intake, physical activity.

Results The distributions of serum 25(OH)D quartiles differed by race/ethnicity, education and smoking status. After adjustment for multiple lifestyle factors, serum 25(OH)D was not related to lean body mass, grip strength or the composite physical function score (all $P > 0{\cdot}20$). There was no variation in the associations between 25(OH)D level and outcomes by race/ethnicity. The relationship between PTH and the outcomes revealed similar results.

Conclusion In this population-based sample of adult men with a broad age range, there was no association between serum 25(OH)D concentration and lean body mass, muscle strength and physical function after controlling for multiple lifestyle factors.

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Correspondence: Andre B. Araujo, Department of Epidemiology, New England Research Institutes, Inc., 9 Galen Street, Watertown, MA 02472, USA. Tel.: 617 972 3152; Fax: 617 926 8246; E-mail: aaraujo@neriscience.com

Introduction

Early case reports of profound vitamin D deficiency and osteomalacia described an associated proximal myopathy, which was reversible by treatment with vitamin D.[1,2] With the identification of the vitamin D receptor in muscle cells[3,4] and neuronal cells,[5] studies in cell culture and experimental animals suggested that vitamin D affects muscle growth, development and contraction.[6] A number of observational studies have shown an association between serum 25-hydroxyvitamin D [25(OH)D] concentration and measures of muscle performance[7–15] and appendicular muscle mass[8,11] in community-dwelling older adults. In addition, several recent randomized controlled trials of vitamin D supplementation report beneficial effects on muscle strength and physical performance in older adults with low vitamin D status.[16–20]

However, evidence for a potential association between serum 25(OH)D level and muscle-related outcomes appears to be strongest in older female-only populations[8,13,15,16,19–21] and older Caucasian populations.[7–9,11–18,20,22] Data examining whether the relationship is consistent in men, particularly younger adult men, are scarce. This is important to know because there is a high prevalence of vitamin D deficiency in men,[23] and after age 40, men lose skeletal muscle mass at a more rapid rate than women.[24]

The aim of this study was to investigate whether serum 25(OH)D concentration was associated with lean body mass, muscle strength and physical performance in a diverse community-dwelling population of adult men.[25] We also examined whether high serum parathyroid hormone (PTH) level was associated with these same muscle outcomes.

Methods

Study sample

Data were obtained from men enrolled in the Boston Area Community Health/Bone (BACH/Bone) Survey, which is a population-based, observational survey study of skeletal health and related outcomes in 1219 (of 1877 eligible, 65% response rate) randomly selected non-Hispanic black, Hispanic and non-Hispanic white male Boston, MA residents aged 30–79 years.[26] Persons of other racial/ethnic backgrounds were not enrolled. BACH/Bone Survey

subjects were a subset of 2301 men previously enrolled in the parent BACH Survey; full details of the BACH survey have been published previously.[27] Study protocols were approved by Institutional Review Boards at New England Research Institutes (NERI) and Boston University School of Medicine (BUSM). All subjects gave written informed consent separately for participation in each study.

Data collection

Interviews and measurements for BACH and BACH/Bone were conducted by trained staff at NERI and the BUSM's General Clinical Research Center (GCRC), respectively. Data collection for BACH generally occurred in subjects' homes. Potential confounders for this analysis included demographic characteristics (age, education, household income), health status (self-reported health, comorbid conditions, family history), hormones [testosterone (T) and sex hormone–binding globulin (SHBG)] and lifestyle [alcohol use, smoking, diet with the Block food frequency questionnaire,[28] and physical activity using the Physical Activity Scale for the Elderly (PASE)].[29] To designate categories of PASE, we calculated quartiles in the entire BACH sample and assigned those scores in the lowest 25% the label 'low', the middle 50% 'middle' and the top 25% 'high'. Self-reported race/ethnicity was determined according to the federal standard.[30] Additional data collected during the BACH/Bone interview included height by stadiometer, weight by digital scale, total body composition by dual energy X-ray absorptiometry (DXA) and a blood draw for measurements such as serum 25(OH)D and bio-intact PTH. Height and weight were then used to calculate body mass index (BMI).

Biochemical measurements

A nonfasting blood sample was collected close to waking time in BACH because of diurnal variation (median time since awakening 3 h 38 min).[31,32] Serum samples were stored at −80 °C until analysis. T and SHBG were measured at the Children's Hospital Medical Center Research Laboratories (Boston, MA, USA) by competitive electrochemiluminescence immunoassays on the 2010 Elecsys system (Roche Diagnostics, Indianapolis, IN, USA). The lower limits of detection for T and SHBG were 0·07 and 3 nmol/l, respectively. The interassay coefficients of variation (CV) for T at concentrations of 0·83–24·31 nmol/l were 7·4–1·7%, 5·2–2·0% and 2·4–2·7% for SHBG at concentrations between 25 and 95 nmol/l. Free T concentrations were calculated from total T and SHBG concentrations using mass action equations.[33,34] Nonfasting serum measures obtained in BACH/Bone included 25(OH)D and bio-intact PTH. These measurements were made throughout the year at the same time as other assessments of interest (i.e., body composition, muscle strength and physical function). Serum 25(OH)D [$25(OH)D_2$ + $25(OH)D_3$] concentration was measured in duplicate (the average of the two are presented) at the Core Laboratory, BUSM, using a competitive binding protein (CPB) assay without prior chromatography.[35] Interassay coefficients of variation are 10–15%. The reference range is 50–250 nmol/l. The lower limit of detection (LLD) for the serum 25(OH)D assay was 12·5 nmol/l; n = 21 men (16 black and 5 Hispanic men) with values less than the LLD were coded to 12·5 nmol/l. PTH was measured in duplicate with the Nichols Advantage System (Nichols Institute Diagnostics, San Clemente, CA, USA) (intra- and interassay CV are 2·2–3·6 and 5·6–8·3%, respectively). The sensitivity for the assay is estimated to be 0·42 pmol/l, and the reference range is 0·84–5·27 pmol/l. The distributions of serum 25(OH)D and PTH were divided into quartiles for analysis. Quartiles of serum 25(OH)D in BACH/Bone were ≤51·9, 52·0–78·0, 78·1–106·6 and ≥106·7 nmol/l. Quartiles of PTH in BACH/Bone were ≤2·14, 2·15–2·88, 2·89–4·06 and ≥4·07 pmol/l.

Measures of physical function and strength

The physical function tests included a timed walking test (time needed to walk 50 ft) and a chair stand test (time needed to stand up and sit down five times with arms folded). Following Guralnik *et al.*,[36] we created a composite 'physical function' variable as follows: those completing the walking test and chair stands were assigned scores of 1–4, corresponding to the quartiles of time needed to complete the test, with the fastest time scored 4. Those who could not complete the test were assigned a score of 0. Thus, the score ranges from 0 to 8. Grip strength of the subject's dominant hand was assessed with a Jamar hydraulic hand dynamometer (Sammons Preston, Bolingbrook, IL, USA), which measures isometric grip force. Subjects were instructed to exert maximum effort for 3 s during two trials, each separated by a 1-min rest. The maximum result was used for analysis.

Measures of body composition

During the BACH/Bone visit, BUSM GCRC staff obtained measurements of nonfat soft tissue (lean mass, LM) from whole-body DXA scans using a QDR 4500W densitometer (Hologic, Inc., Waltham, MA, USA). All mass quantities reported here exclude the head. LM was calculated by subtracting bone mineral mass from nonfat mass. The DXA system was monitored weekly for drift.

Analytic sample

Of the 1209 subjects that completed lower body physical function tests, we excluded 94 subjects who had 25(OH)D values that were missing (n = 84) or were considered outliers (>200 nmol/l) (n = 10). This left 1115 men as a base analysis sample. More specific analysis samples were developed based on missing data on covariates that were relevant to the final models. From the base sample, three subjects did not complete the PASE questionnaire, five did not report arthritis status, one was missing alcohol usage, 66 were missing education or income and 142 men did not provide data on calcium consumption. Final models exploring lower body physical function were reduced to N = 898. Grip strength final models were reduced to N = 753, and LM models examined a total of N = 852 men.

Statistical methods

To account for the complex sampling design, weighted analyses were conducted using SUDAAN 9.0.1 (Research Triangle Institute, Research Triangle Park, NC, USA). Descriptive data were presented on all outcomes and potential confounding and explanatory variables by 25(OH)D quartiles. Differences by 25(OH)D quartiles were tested using chi-square tests for categorical variables and Wald-type *F* tests for continuous variables.

Exploratory analyses investigating whether the relationship between serum 25(OH)D level and outcomes varied by race/ethnicity were performed. This included LOESS plots of the outcomes *vs* serum 25(OH)D level, regression models testing the significance of interaction terms between 25(OH)D and race/ethnicity, and racial/ethnic group-specific models regressing outcomes on 25(OH)D level. Analyses examining whether the relationship between serum 25(OH)D level and outcomes varied by age (age <65 *vs* ≥65 years) were also performed.

Least-square means were used to describe the relationship between 25(OH)D quartiles and outcomes, adjusted for confounding influences. Four sets of regression models were constructed: unadjusted, age adjusted, age and race/ethnicity adjusted, and multivariate adjusted. Backwards stepwise elimination was used for variable selection in the multivariate-adjusted models to reach the most parsimonious final model (using $P < 0{\cdot}05$). Final models were run adjusting for all covariates that were significant in any of the three multivariate-adjusted models. Final multivariate models were adjusted for age, race/ethnicity, calcium intake, PASE score, BMI, education, income, self-reported health, arthritis and alcohol.

We ran crude as well as adjusted correlations between continuous 25(OH)D and outcomes. These analyses were repeated for the relationship between PTH and outcomes.

Results

There were no statistically significant findings from regression models testing whether the relationship between 25(OH)D and the outcomes varied by race/ethnicity or by age group, nor did exploratory LOESS plots reveal any variation by race/ethnicity or age group, so subsequent analyses were performed in the total sample.

Table 1 describes the analysis sample by serum 25(OH)D quartiles. Subjects varied significantly by serum 25(OH)D quartile in race, education, smoking status and serum PTH level. As previously reported,[25] the lowest serum 25(OH)D quartile had a higher proportion of black men and current smokers than the other three quartiles (Table 1). In the entire sample, the average (±SD) LM was 55·2 ± 7·8 kg and the mean maximum grip strength was approximately 40 ± 12 kg. Physical composite scores ranged from 0 to 8 with a mean of 5·0 ± 2·0. In the sample, 19·1% had 25(OH)D levels <50 nmol/l and 42·2% had 25(OH)D levels <75 nmol/l. In analyses not shown, subjects included in the analysis samples differed significantly from the excluded subjects with regard to race/ethnicity, socioeconomic status and physical activity; minorities, men of lower socioeconomic status and less physically active men were more likely to be excluded.

Preliminary analysis indicated linear associations between 25(OH)D and all outcomes. The crude and adjusted means of outcome variables according to quartiles of serum 25(OH)D are presented in Table 2. In most cases, outcomes were similar across quartiles of serum 25(OH)D (all $P > 0{\cdot}20$). In an age-adjusted model, serum 25(OH)D was related to the composite physical function score ($P = 0{\cdot}03$). However, in racial/ethnic- and multivariate-adjusted models, serum 25(OH)D was no longer related to physical function score (Table 2). Parallel correlation analyses treating serum 25(OH)D as a continuous variable also demonstrated no significant relationship between serum 25(OH)D and the outcomes after adjustment for race/ethnicity (results not shown). When adjusted for season, results were unchanged. Analysis examining the relationship between PTH and the outcomes revealed similar nonsignificant results (results not shown).

Discussion

In this population-based sample of adult men, we did not find a significant association between serum 25(OH)D concentration and lean body mass, hand grip strength or physical function (walking speed and chair stand) after careful adjustment for health and lifestyle factors, including physical activity. Notably, men in the lowest 25(OH)D quartile appeared to differ significantly from those in higher quartiles by way of a greater percentage of blacks and current smokers.

Our findings are not consistent with a large number of cross-sectional studies on vitamin D status and muscle performance;[7–10,13,14,22,37] however, there are several distinguishing aspects of our study that may account for these differences. First, our study population consisted of only adult men. Most published research on vitamin D and its association with physical performance has either been conducted in samples of women or a mixed sample. Some of the observational studies involving both women and men have reported sex-specific findings. In an analysis of older adults who participated in the observational Rancho Bernardo Study, performance on a timed up and go test and chair stand was poorer and declined at a faster rate over a 2·5-year period in women with 25(OH)D levels ≤80 nmol/l compared to women with 25(OH)D levels ≥115 nmol/l.[38] This finding was not seen in older male participants in this study. In 337 older adults, skeletal muscle mass correlated significantly with levels of 25(OH)D in women only, but not in the men.[39] Interestingly, a single randomized trial involving older men by Kenny *et al.*[40] reported that vitamin D_3 1000 IU plus calcium 500 mg daily, *vs* calcium alone, did not increase muscle strength or improve physical performance over a 6-month period. Thus, these data raise the possibility that vitamin D status is more commonly associated with muscle performance in women rather than men. Reasons for this sex difference are uncertain, yet potential explanations may involve differences in baseline lean body mass, muscle strength, physical activity and circulating levels of testosterone, a hormone that is anabolic to muscle.[41,42]

Another distinctive aspect of our study was the broad age range of the participants. Prior population-based studies[7,9–13,15] and intervention studies[16–20] showing positive results included

Table 1. Descriptive characteristics of the study population by serum 25(OH)D quartile

	Overall Mean ± SD or %	25(OH)D quartile 1 (≤51·9 nmol/l) Mean ± SD or %	25(OH)D quartile 2 (52·0–78·0 nmol/l) Mean ± SD or %	25(OH)D quartile 3 (78·1–106·6 nmol/l) Mean ± SD or %	25(OH)D quartile 4 (≥106·7 nmol/l) Mean ± SD or %	*P*-value*
Age, years	47·9 ± 12·8	48·1 ± 12·8	47·4 ± 12·8	47·7 ± 12·7	48·3 ± 13·0	0·95
Race/ethnicity (%)						
Black	25·0	52·8	23·9	17·2	11·6	<0·001
Hispanic	12·5	13·9	11·1	15·2	10·3	
White	62·5	33·3	65·0	67·7	78·0	
BMI, kg/m^2	28·5 ± 4·7	29·0 ± 5·6	28·0 ± 4·0	28·5 ± 4·5	28·5 ± 4·8	0·22
Calcium intake/day, mmol	21·3 ± 14·5	19·9 ± 18·3	21·5 ± 14·2	21·4 ± 13·5	22·0 ± 12·5	0·89
PASE (%)						
Low	24·5	29·7	28·6	16·9	23·7	0·11
Middle	47·7	41·9	46·0	53·7	48·3	
High	27·8	28·4	25·5	29·4	28·0	
Household income, $ (%)						
<10 000	14·2	24·0	13·2	10·7	10·8	0·06
10 000–29 999	22·8	24·1	21·8	21·0	24·5	
30 000–69 999	32·7	25·2	39·8	28·5	35·9	
70 000+	30·4	26·7	25·2	39·8	28·8	
Education, years	15·2 ± 4·1	14·2 ± 4·1	15·7 ± 4·3	15·7 ± 3·8	15·0 ± 4·1	0·01
Self-reported health (%)						
Excellent	20·4	14·9	21·1	23·6	20·9	0·12
Very good	34·9	28·0	35·1	38·5	36·6	
Good	30·3	37·2	27·7	27·0	30·6	
Fair or poor	14·4	19·8	16·0	10·9	11·9	
Number of major comorbidities (%)†						
0	47·0	42·3	46·4	48·6	49·6	0·17
1	33·7	34·8	34·2	35·1	31·0	
2	12·0	14·1	7·7	11·9	14·5	
3+	7·4	8·8	11·7	4·3	4·9	
Arthritis (% yes)	22·2	24·2	19·2	23·2	22·6	0·76
Smoking status (%)						
Never	45·0	34·3	43·0	53·2	47·7	0·002
Former	28·9	24·7	29·3	29·7	30·9	
Current	26·1	40·9	27·7	17·1	21·4	
Alcohol, drinks/day (%)						
0	25·0	27·0	28·3	22·3	22·7	0·46
<1	38·4	33·1	37·7	41·7	40·1	
1–3	27·2	23·8	27·2	26·8	30·3	
>3	9·4	16·1	6·8	9·3	6·9	
Total testosterone, nmol/l	15·2 ± 6·5	15·6 ± 7·7	15·9 ± 6·2	15·1 ± 5·8	14·2 ± 6·3	0·31
Free testosterone, nmol/l	0·31 ± 0·12	0·33 ± 0·13	0·33 ± 0·13	0·31 ± 0·11	0·29 ± 0·13	0·18
PTH, pmol/l	3·27 ± 1·95	3·70 ± 2·54	3·37 ± 1·62	3·25 ± 2·01	2·87 ± 1·56	0·003

25(OH)D, 25-hydroxyvitamin D; PASE, Physical Activity Scale for the Elderly; PTH, parathyroid hormone.
**P*-value testing the null hypothesis of no difference by 25(OH)D quartile in the variable under consideration.
†Major comorbidities include: asthma, chronic lung disease, myocardial infarction, stroke, high cholesterol and high blood pressure.

individuals of at least age 65 or older, community-dwelling or institutionalized. Studies in a mixed-age adult population of younger and older women[43] and in younger postmenopausal women (early 60s)[44] did not find a correlation between vitamin D status and tests of muscle performance. Furthermore, given that our study population is younger and community-dwelling, our subjects have an overall higher level of physical functioning compared to a more aged population; this may result in a 'ceiling effect' when attempting to examine associations between physical function and a potential predictor such as 25(OH)D level.[45,46] Our sample data, however, did not reveal an interaction effect between age group [grouped younger <65 years *vs* older ≥65 years (N = 180)] and serum 25(OH)D concentration on the muscle outcomes. Nonetheless, one may speculate that those who stand

Table 2. Least squares means of physical performance measures and lean mass according to serum 25(OH)D quartiles

25(OH)D quartile	Crude	Age-adjusted	Age- and racial/ethnic group-adjusted	Multivariate-adjusted*
Composite physical function score (N = 898)				
Quartile 1 (≤51·9 nmol/l)	4·49	4·50	4·73	4·79
Quartile 2 (52·0–78·0 nmol/l)	5·14	5·11	5·09	5·00
Quartile 3 (78·1–106·6 nmol/l)	5·17	5·16	5·12	4·96
Quartile 4 (≥106·7 nmol/l)	5·08	5·10	4·98	5·02
P-value†	0·07	0·03	0·44	0·76
Grip strength (N = 753)				
Quartile 1 (≤51·9 nmol/l)	38·9	39·1	38·7	38·6
Quartile 2 (52·0–78·0 nmol/l)	38·8	38·9	38·8	38·9
Quartile 3 (78·1–106·6 nmol/l)	42·5	42·3	42·5	41·8
Quartile 4 (≥106·7 nmol/l)	39·2	39·3	39·4	39·1
P-value†	0·24	0·28	0·22	0·29
Lean mass (N = 852)				
Quartile 1 (≤51·9 nmol/l)	55·1	55·1	54·9	54·4
Quartile 2 (52·0–78·0 nmol/l)	54·8	54·7	54·6	55·1
Quartile 3 (78·1–106·6 nmol/l)	55·9	55·9	56·1	55·6
Quartile 4 (≥106·7 nmol/l)	55·2	55·2	55·3	54·8
P-value†	0·62	0·56	0·36	0·29

25(OH)D, 25-hydroxyvitamin D; PASE, Physical Activity Scale for the Elderly; BMI, body mass index.
*After backwards stepwise elimination for models ($P > 0·05$), remaining covariates from any of the three models were controlled for in all three final models. Age and race/ethnicity remained in the multivariate-adjusted models regardless of significance. Final multivariate models were adjusted for: calcium intake, PASE score, BMI, education, income, self-reported health, arthritis and alcohol intake.
†P-value testing the null hypothesis of no difference in the variable under consideration by 25(OH)D quartile.

to benefit most from vitamin D supplementation may be an older population with evidence of physical impairment and vitamin D insufficiency.

Lastly, our study had a higher proportion of individuals of non-Caucasian race than most prior studies. We, however, saw no evidence that the association of 25(OH)D level with outcome measures differed across race/ethnic groups. In this population-based group of adult men, PTH concentration was not associated with muscle strength or physical function. A few studies have reported an association in a sample of older men and women,[11,47] but not in younger age groups.

The strengths of this study include its random, population-based sample with a large number of men across a broad age range with robust measurement of variables of interest. Hand grip strength[48,49] has been validated in young adults, and the chair stand test[50] has been validated in individuals over age 55; however, to our knowledge, the walk test used has not been validated in younger adults. In addition, the majority of the men in this study sample had serum 25(OH)D levels below a commonly accepted target level of 75 nmol/l,[51] especially in view of the CPB 25(OH)D assay, which generally overestimates serum 25(OH)D levels by 15–20%.[25] Thus, these findings do not preclude that higher 25(OH)D levels, as would be achieved through vitamin D supplementation, might provide benefit. This study is limited by the fact that minorities, men of lower socioeconomic status and less physically active men were more likely to be excluded from the analysis sample.

In conclusion, in this diverse, population-based sample of adult men of varying ages, there was no association between serum 25-hydroxyvitamin D concentration and lean body mass, muscle strength or physical function after controlling for multiple health and lifestyle factors. Further investigation into additional diverse populations of men is needed to confirm these findings.

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Disclosure

The authors have nothing to declare.

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