Association of Plasma Vitamin D Levels with Adiposity in Hispanic and African Americans

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Context: Previous studies have suggested vitamin D insufficiency is associated with increased obesity; however, the relationship between 25-hydroxyvitamin D (25[OH]D) and 1,25-dihydroxyvitamin D ($1,25[OH]_2D$) and measures of adiposity has not been well characterized in minority populations.

Objective: The objective of the study was to examine the relationship between levels of 25[OH]D and $1,25[OH]_2D$ and measures of adiposity in Hispanic and African-Americans at baseline and on change in these measures over time.

Design and Setting: The Insulin Resistance Atherosclerosis (IRAS) Family Study examined 917 Hispanics and 439 African-Americans at baseline and again 5.3 yr later (n = 1081 at follow-up).

Main Outcome Measure: 25[OH]D (nanograms per milliliter) and $1,25[OH]_2D$ (picograms per milliliter) were measured at baseline. Abdominal sc adipose tissue (SAT), visceral adipose tissue (VAT; both determined by computed tomography scan), and body mass index (BMI) were measured at baseline and follow-up.

Results: 25[OH]D was inversely associated with BMI, VAT, and SAT in both populations at baseline ($P < 0.001$). 25[OH]D was marginally inversely associated with baseline visceral fat to sc fat ratio in African-Americans ($P = 0.049$) but not Hispanics. $1,25[OH]_2D$ was inversely associated with BMI ($P < 0.0001$, $P = 0.002$) and VAT ($P = 0.0005$, $P = 0.012$) in Hispanics and African-Americans, respectively, whereas $1,25[OH]_2D$ was inversely associated with SAT in Hispanics ($P < 0.0001$) and with visceral fat to sc fat ratio in African-Americans ($P = 0.02$). Adjusting for 25[OH]D attenuated these associations; $1,25[OH]_2D$ remained associated with BMI in both populations ($P < 0.05$) and with SAT ($P = 0.004$) in Hispanics. No significant associations between 5-yr change in adiposity and 25[OH]D or $1,25[OH]_2D$ were seen.

Conclusions: Vitamin D levels were inversely associated with baseline BMI, SAT, and VAT in Hispanic and African-Americans but were not associated with 5-yr change in adiposity. **(*J Clin Endocrinol Metab* 94: 3306–3313, 2009)**

Lower levels of 25-hydroxyvitamin D (25[OH]D) have been associated with glucose intolerance, altered insulin secretion and type 2 diabetes (1). Vitamin D deficiency is also emerging as a risk factor for the metabolic syndrome in adults (2). Current literature supports an inverse relationship between 25[OH]D and components of the metabolic syndrome, including high blood glucose concentration, insulin resistance,

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Abbreviations: BMI, Body mass index; CT, computed tomography; EEE, estimated energy expenditure; FSIGT, frequently sampled iv glucose tolerance test; IRAS, Insulin Resistance Atherosclerosis; MET, metabolic equivalent; 25[OH]D, 25-hydroxyvitamin D; $1,25[OH]_2D$, 1,25-dihydroxyvitamin; SAT, sc adipose tissue; VAT, visceral adipose tissue; VSR, visceral fat to sc fat ratio.

dyslipidemia, elevated blood pressure, and abdominal obesity (2).

Obesity, which is a known risk factor for these chronic conditions, has been, on its own, associated with vitamin D deficiency in both adults (3–18) and children and adolescents (7, 19). Increased storage of 25[OH]D in adipose tissue is a plausible explanation for increased rates of vitamin D deficiency in obese individuals (16) because 25[OH]D level is inversely associated with total body fat (13). However, obese individuals may be at increased risk for vitamin D deficiency due to decreased sun exposure from increased clothing or limited mobility (5, 15), although this has not been found in all studies (17). Alternatively, obesity may also be the consequence of low vitamin D levels because it has been hypothesized that low vitamin D status, by causing PTH excess and calcium influx into adipocytes, may promote weight gain (20). In addition, 1,25-dihydroxyvitamin D (1,25$[OH]_2$D), which is derived from 25[OH]D, may inhibit adipogenesis (21).

Whereas previous studies have provided a link between adiposity measures and vitamin D, these measures have largely been limited to body mass index (BMI) (3–16, 18, 19), waist circumference (8, 13, 14), waist to hip ratio (13, 14), and fat mass as measured by bioelectrical impedance (14, 19) and dual-energy x-ray absorptiometry scan (11, 13, 15, 17). These studies can examine the general association of fat and obesity with vitamin D; however, it is known that the type of fat is important in determining risk for disease. For instance, higher visceral fat is associated with increased risk of diabetes, heart attack, and stroke (22). Visceral and sc fat was associated with 25[OH]D levels in a small study of 53 Hispanic and 37 Caucasian young women (23). More data are needed in larger minority populations of both genders and in older age groups to further understand the relationship between location of fat and vitamin D. In a large population of adults between the ages of 18 and 81 yr, we examined computed tomography (CT)-derived measures of sc adipose tissue (SAT), visceral adipose tissue (VAT) and visceral fat to sc fat ratio (VSR; a measure of the distribution, rather than quantity, of fat), along with 25[OH]D and 1,25$[OH]_2$D in Hispanic-American and African-American men and women who are at increased risk for obesity as well as vitamin D deficiency. Levels of 25[OH]D in the Insulin Resistance Atherosclerosis (IRAS) Family Study have been shown to be highest in Hispanics from San Luis Valley, CO, lower in Hispanics from San Antonio, TX, and lowest in African-Americans from Los Angeles, CA, with means between 11.0 ng/ml and 18.3 ng/ml (24). This indicates this population would be beneficial to study because it is at risk of being vitamin D deficient (25[OH]D levels $<$20 ng/ml).

Subjects and Methods

The IRAS Family Study was designed to explore genetic and epidemiological contributions to abdominal adiposity and glucose homeostasis traits among Hispanic- and African-Americans using a family-based design (25). In the IRAS Family Study, family members of the original IRAS participants (1625 unrelated individuals were examined to determine the relationship between insulin resistance and atherosclerosis) were recruited to participate in a baseline clinical examination between 1999 and 2002. Additional families were recruited from the general population to supplement the IRAS families. Ascertainment and recruitment of families were based on family size and not on phenotype. Hispanic families were recruited from San Antonio, TX, and the San Luis Valley, CO. African-American families were recruited from Los Angeles, CA. A follow-up examination was conducted approximately 5 yr after the baseline examination (mean follow-up 5.3 yr). The institutional review boards at the respective institutions approved the protocol, and informed consent was given by each subject.

Identical protocols were followed for the baseline and follow-up visits. Height and weight were measured to the nearest 0.5 cm and 0.1 kg, respectively. BMI was calculated as weight (kilograms)/height (square meters). Abdominal fat mass was measured at the L2/L3 and L4/L5 vertebral regions by CT under a common protocol at each of the three sites. Before both the baseline and follow-up phases of the study, a training session was conducted for each site by a centralized CT Reading Center at the University of Colorado Denver. Sites were certified when they had passed the training and when acceptable sample images had been successfully transferred to the CT Reading Center. During the course of the study, images were continually reviewed for the image quality and imaging technique and rescans were performed when needed.

The effective whole-body radiation dose for the images done in this study did not exceed 100 mrem. Exclusions for the CT scan included the inability to lay supine, weight exceeding the limit for the CT table (generally 350–400 lb) and pregnancy. All subjects were gowned and all tight undergarments, including underwear, were removed. Patients were placed in a supine position with the feet or head directed toward the gantry and with the arms above the head. Care was taken to position the patient symmetrically on the CT table. No pads or cushions, other than the standard table pad, were used.

All patients received an anterior-posterior scout of the abdomen and pelvis (diaphragm through symphysis pubis) followed by three axial images. Optimal parameters for the scout varied with CT model and patient body habitus. After the scout was obtained, the L4-L5 disk space was located by counting the lumbar vertebra with L1 being the first non-rib-bearing vertebra. In the unusual event that there were more or less than five non-rib-bearing lumbar vertebrae, the disk space closest to the iliac crest was considered to be L4-L5. The L2-L3 disk was identified as the second one above L4-L5. A single 10-mm-thick image was obtained through the L2-L3 disk space, followed by a single 10-mm-thick axial image through the L4-L5 disk space, both during suspended respiration.

Both the baseline and follow-up CT images were read centrally at the CT Reading Center at the University of Colorado Denver. The CT scans were analyzed using IDL version 6.3 software (Research Systems, Inc, Boulder, CO). The areas of VAT and SAT were calculated. VAT and SAT at the L4/L5 region were

chosen for these analyses. For those subjects on whom we had data for the L2/L3 region but not the L4/L5 region, fat areas in the L4/L5 region were imputed from those in the L2/L3 region using a simple linear model because adipose tissue areas at the L2/L3 and L4/L5 regions were highly correlated (Spearman correlation: 0.95 for SAT, 0.90 for VAT). VSR was calculated as the VAT at L4-L5/SAT at L4/L5. Imputed L4/L5 VAT or SAT data were not used to calculate baseline VSR, thus resulting in a slightly reduced study sample for this variable. For the change in adiposity phenotypes, we excluded those subjects who did not have L4/L5 readings at both time points.

Levels of 25[OH]D were measured by a two-step process involving rapid extraction of 25[OH]D and other hydroxylated metabolites from plasma and RIA with a 25[OH]D-specific antibody (DiaSorin, Stillwater, MN) with interassay coefficients of variation less than 8%. Levels of $1,25[OH]_2D$ were measured by a two-step process involving extraction and purification of vitamin D metabolites from plasma and RIA with a $1,25[OH]_2D$-specific antibody (DiaSorin) with interassay coefficients of variation less than 19%. Season of blood draw was determined from the date of the baseline visit. Blood draws taken from December through May were categorized as winter/spring and those taken from June through November were categorized as summer/fall (24).

Physical activity was assessed by a 1-yr recall using a modification of a validated instrument (26) that incorporated activities common among IRAS Family Study participants, including ranching and homemaking activities. These activities were queried in groups according to home, work, or leisure time and according to intensity of activities (light, moderate, or vigorous) based on metabolic equivalent (MET) values. For each activity group, usual frequency and duration of participation was recorded, from which estimated energy expenditure (EEE) was determined. Total energy expended (in kilocalories per kilogram) per year was calculated by summing across all activity groups, plus the EEE from sleep (MET value of 1.0), plus the EEE from light activities (*e.g.* sitting MET value of 1.5). We analyzed two physical activity variables: energy expenditure per year from all activities and energy expenditure per year from vigorous activities.

Data on smoking habits, marital status, and education level were gathered by standardized questionnaire (25).

A total of 1748 participants attended the baseline visit of the IRAS Family Study and had an abdominal CT scan. Of these, 207 were classified as diabetic and were excluded from this analysis. Of the remaining 1541 participants, 1356 had a plasma sample from a frequently sampled iv glucose tolerance test (FSIGT) done at the baseline visit that could be used for vitamin D testing. Reasons for not having an FSIGT at the baseline visit included having an FSIGT within the past 5 yr for a different study protocol, medical contraindications, iv line failure, and participant refusal. The mean BMI of those with and without vitamin D was similar (29.1 and 28.4, respectively, $P = 0.14$). Of the 1356 with baseline vitamin D and CT scan data, 946 participants had a repeat CT scan at the IRAS Family Study follow-up visit and therefore had 5-yr adiposity change data available for analysis.

Statistical methods

For all adiposity phenotypes (baseline and change), extreme outliers were verified and were then winsorized (*i.e.* truncated to the next largest value present in the data) before analyses. Distributions of BMI, VAT, SAT, and VSR levels were positively skewed at baseline. Therefore, a square root transformation was used to better approximate a normal distribution for VAT and SAT, whereas a log transformation was used for BMI and VSR. Spearman correlations by gender and ethnicity indicate a high collinearity between adiposity phenotypes: correlations between BMI and SAT ranged from $r^2 = 0.89$ to 0.92, between BMI and VAT ranged from $r^2 = 0.59$ to 0.70, and between VAT and SAT ranged from $r^2 = 0.61$ to 0.65 (27). For each phenotype, change in adiposity was calculated as yr 5 follow-up minus baseline. No transformation of the change data were needed. Based on observations that the relationship between age and our adiposity change phenotypes was nonlinear (28) we used age and age^2 in these models to account for this nonlinear relationship.

Variance component analysis implemented in SOLAR was used to examine associations and account for the correlations among family members in pedigrees of arbitrary size and complexity (29). Statistical details of the application of this approach are described by Kammerer *et al.* (30). Whereas gender differences in adiposity composition exist, our primary goal was to examine whether differences in adiposity between Hispanics and African-Americans could be predicted by the differences seen in the plasma vitamin D levels of these two ethnic groups. Therefore, we ran our models separately by ethnicity and adjusted for gender. All models were adjusted for age and gender, and the models in Hispanics were also adjusted for clinic site (San Antonio and San Luis Valley). Additional variables were included in the model if they were statistically significant risk factors of the adiposity trait. In each phenotype (BMI, VAT, SAT, VSR), if a variable was significant for one ethnic group, it was included in the model for the other ethnic group, regardless of statistical significance, for ease of comparison across ethnic groups. The significance of each variable was assessed by the likelihood ratio test. To determine the relative amount of the variance in the outcome explained by vitamin D level, the final models were run with and without vitamin D as a predictor variable.

Results

Characteristics of the participants at baseline and of those for whom we have 5-yr change data are shown in Table 1. At baseline, the mean 25[OH] level was 16.6 ng/ml for Hispanics and 11.0 ng/ml for African-Americans. In both Hispanics and African-Americans, males had higher 25[OH]D and $1,25[OH]_2D$ than females. In general, males had more visceral fat, whereas females had more sc fat at both baseline and follow-up.

Baseline analyses

Lower 25[OH]D level was significantly associated with BMI in both the Hispanic and African-American populations at baseline, adjusting for age, sex, and smoking status (Table 2). The proportion of variance explained by vitamin D level was 0.05 in Hispanics, which was 50% of the total variance explained by the model. In African-Americans, the proportion of variance explained by 25[OH]D was 0.02, which was 33.3% of the total variance explained by the model.

TABLE 1. Characteristics of IRAS Family Study participants in the baseline adiposity analyses and the change in adiposity analyses

	Population used in baseline analyses				Population used in change analyses[a]			
	Hispanics (n = 917)		African-Americans (n = 439)		Hispanics (n = 657)		African-Americans (n = 289)	
Characteristic	Male (n = 380)	Female (n = 537)	Male (n = 191)	Female (n = 248)	Male (n = 256)	Female (n = 401)	Male (n = 120)	Female (n = 169)
Age at clinic visit, mean ± SD (median)[b]	38.4 ± 13.4 (35.6)	39.9 ± 12.6 (39.7)	42.1 ± 14.3 (40.3)	39.5 ± 12.7 (38.8)	43.7 ± 13.4 (41.0)	44.6 ± 12.1 (45.0)	48.6 ± 14.6 (46.5)	46.1 ± 12.4 (45.5)
25[OH]D at baseline (ng/ml), mean ± SD (median)[c]	17.9 ± 6.7 (18.0)	15.7 ± 7.7 (14.0)	12.1 ± 5.6 (11.0)	10.2 ± 5.2 (8.8)	18.3 ± 5.2 (18.0)	15.6 ± 7.4 (14.0)	12.0 ± 5.6 (11.5)	10.5 ± 5.1 (8.9)
$1,25[OH]_2D$ at baseline (pg/ml), mean ± SD (median)[d]	47.2 ± 14.6 (46.0)	45.6 ± 16.1 (43.0)	44.8 ± 13.4 (44.5)	42.9 ± 14.1 (41.0)	47.7 ± 5.2 (46.0)	45.0 ± 14.9 (43.0)	45.7 ± 13.7 (46.0)	43.8 ± 14.0 (42.0)
Current smoker at baseline								
Yes	111 (29.2%)	109 (20.2%)	51 (26.7%)	53 (21.4%)	64 (25.0%)	75 (18.7%)	25 (20.8%)	39 (23.1%)
No	269 (70.8%)	428 (79.8%)	140 (73.3%)	195 (78.6%)	192 (75.0%)	326 (81.3%)	95 (79.2%)	130 (76.9%)
Marital status at baseline[e]								
Unmarried	177 (46.7%)	252 (47.0%)	105 (55.0%)	160 (64.5%)	112 (43.9%)	188 (46.9%)	63 (52.5%)	107 (63.3%)
Married	202 (53.3%)	284 (53.0%)	86 (45.0%)	88 (35.5%)	144 (56.1%)	213 (53.1%)	57 (47.5%)	62 (36.7%)
Total EEE at baseline (kcal/kg · yr), mean ± SD (median)	18082.6 ± 4789.4 (17076.6)	15460.3 ± 3010.4 (14624.6)	16058.9 ± 4115.2 (14627.3)	14786.3 ± 2928.3 (13742.9)	18461.7 ± 5502.7 (16653.6)	15685.4 ± 3623.3 (14444.0)	16656.9 ± 4501.6 (14734.3)	15169.1 ± 3247.6 (14108.6)
EEE from vigorous activity at baseline (kcal/kg · yr), mean ± SD (median)	4113.9 ± 4727.1 (2152.0)	1031.6 ± 1814.3 (352)	3012.7 ± 4048.1 (1672.0)	1005.9 ± 1713.3 (388.0)	4739.1 ± 5423.7 (2354.0)	1492.6 ± 2476.9 (536.0)	3589.3 ± 4335.2 (1660.0)	1277.5 ± 2174.8 (512)
Education at baseline[f]								
More than high school	149 (40.2%)	200 (38.7%)	69 (39.0%)	79 (33.5%)	112 (44.6%)	146 (38.1%)	28 (25.7%)	48 (30.4%)
High school or less	222 (59.8%)	313 (61.3%)	108 (61.0%)	157 (66.5%)	139 (55.4%)	237 (61.9%)	81 (74.3%)	110 (69.6%)
Season of baseline blood draw								
Summer	158 (41.7%)	216 (40.2%)	93 (48.4%)	101 (40.9%)	113 (44.3%)	152 (37.7%)	61 (51.3%)	75 (44.4%)
Winter	222 (58.3%)	321 (60.8%	98 (51.6%)	147 (59.1%)	143 (55.7%)	249 (62.3%)	59 (48.7%)	94 (55.6%)
Currently employed at baseline[e]								
Yes	328 (86.3%)	487 (90.5%)	165 (86.4%)	210 (84.7%)	228 (89.0%)	369 (92.0%)	106 (88.3%)	138 (81.7%)
No	52 (13.7%)	48 (8.5%)	26 (13.6%)	38 (15.3%)	28 (11.0%)	32 (8.0%)	14 (11.7%)	31 (18.3%)
BMI (kg/m^2), mean ± SD (median)[b]	27.9 ± 5.1 (27.7)	28.6 ± 6.3 (27.5)	28.4 ± 5.2 (28.0)	30.0 ± 7.4 (28.7)	28.6 ± 5.0 (28.1)	29.3 ± 6.3 (28.4)	29.3 ± 4.9 (28.6)	32.2 ± 8.3 (31.8)
SAT (cm^2), mean ± SD (median)[b]	264 ± 128 (243)	375 ± 148 (352)	253 ± 140 (233)	404 ± 193 (378)	282 ± 132 (256)	404 ± 53 (381)	299 ± 158 (278)	452 ± 209 (430)
VAT (cm^2), mean ± SD (median)[b]	121 ± 60 (113)	97 ± 53 (87)	97 ± 61 (86)	77 ± 47 (67)	123 ± 56 (113)	95 ± 154 (86)	106 ± 60 (99)	90 ± 55 (80)
VSR, mean ± SD (median)[b]	0.5 ± 0.2 (0.5)	0.3 ± 0.1 (0.2)	0.4 ± 0.2 (0.4)	0.2 ± 0.1 (0.2)	0.5 ± 0.2 (0.4)	0.2 ± 0.1 (0.2)	0.4 ± 0.2 (0.3)	0.2 ± 0.09 (0.2)
Change in BMI (kg/m^2), mean ± SD (median)	N/A	N/A	N/A	N/A	0.8 ± 2.2 (0.7)	0.9 ± 2.8 (0.9)	1.1 ± 2.4 (0.9)	2.0 ± 3.4 (1.9)
Change in SAT (cm^2), mean ± SD (median)	N/A	N/A	N/A	N/A	25 ± 55 (18)	25 ± 75 (23)	35 ± 65 (20)	40 ± 86 (45)
Change in VAT (cm^2), mean ± SD (median)	N/A	N/A	N/A	N/A	4 ± 36 (7)	2 ± 30 (2)	3 ± 36 (2)	7 ± 28 (9)
Change in VSR, mean ± SD (median)	N/A	N/A	N/A	N/A	−0.02 ± 0.1 (−0.02)	−0.01 ± 0.07 (−0.01)	−0.03 ± 0.1 (−0.02)	−0.003 ± 0.05 (−0.0001)

N/A, Not applicable.

[a] Reflects the number of IRAS Family Study participants in the baseline analyses who also came in for a follow-up visit an average of 5.3 yr later.

[b] The age, BMI, VAT, SAT, and VSR values displayed in the Change in Adiposity Population columns are those measured at the follow-up visit.

[c] Multiply by 2.496 to convert to the International System of Units.

[d] Multiply by 2.6 to convert to the International System of Units.

[e] Marital status and current employment is missing for two Hispanic individuals at baseline.

[f] Education is missing for 33 Hispanics and 26 African-Americans at baseline and 23 Hispanic and 22 African-Americans for the change analyses.

TABLE 2. Regression coefficients (± SE) from multivariable models of factors associated with adiposity phenotypes at the IRAS Family Study baseline visit

	Adiposity phenotype							
	BMI Covariate coefficients ± SE *P* value		SAT Covariate coefficients ± SE *P* value		VAT Covariate coefficients ± SE *P* value		VSR Covariate coefficients ± SE *P* value	
Baseline covariate	Hispanics (n = 917)[a]	African-Americans (n = 439)	Hispanics (n = 912)	African-Americans (n = 431)	Hispanics (n = 913)	African-Americans (n = 439)	Hispanics (n = 888)	African-Americans (n = 427)
Age	0.002 ± 0.0005	0.004 ± 0.0008	0.02 ± 0.01	0.08 ± 0.02	0.10 ± 0.006	0.12 ± 0.008	0.02 ± 0.0009	0.02 ± 0.001
	$P = 0.0002$	$P < 0.0001$	$P = 0.049$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$
Sex (female)	−0.01 ± 0.01	0.05 ± 0.02	2.57 ± 0.27	4.36 ± 0.48	−1.53 ± 0.15	−0.73 ± 0.21	−0.67 ± 0.02	−0.68 ± 0.04
	$P = 0.35$	$P = 0.01$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P = 0.0006$	$P < 0.0001$	$P < 0.0001$
25[OH]D	−0.006 ± 0.0009	−0.006 ± 0.002	−0.13 ± 0.02	−0.11 ± 0.04	−0.06 ± 0.19	−0.09 ± 0.02	−7.50E-07 ± 0.0018	−0.007 ± 0.004
	$P < 0.0001$	$P = 0.0009$	$P < 0.0001$	$P = 0.007$	$P < 0.0001$	$P < 0.0001$	$P = 0.99$	$P = 0.049$
Current smoker	−0.06 ± 0.01	−0.05 ± 0.02	−1.03 ± 0.30	−0.59 ± 0.52	[b]	[b]	−0.07 ± 0.03	−0.06 ± 0.04
	$P = 0.0001$	$P = 0.02$	$P = 0.0005$	$P = 0.25$			$P = 0.02$	$P = 0.20$
Currently unmarried	[b]	[b]	−0.62 ± 0.25	−0.19 ± 0.47	−0.40 ± 0.15	−0.31 ± 0.22	[b]	[b]
			$P = 0.01$	$P = 0.68$	$P = 0.011$	$P = 0.16$		
EEE from vigorous activity (kcal/kg · yr)	[b]	[b]	$-6 \times 10^{-5} \pm 3 \times 10^{-5}$	$-7 \times 10^{-5} \pm 7 \times 10^{-5}$	[b]	[b]	[b]	[b]
			$P = 0.03$	$P = 0.36$				
Variance explained by model	0.10	0.06	0.22	0.21	0.31	0.41	0.54	0.56
Variance explained by 25[OH]D	0.05	0.02	0.05	0.02	0.03	0.03	<0.01	0.006

[a] Models in the Hispanic population were also adjusted for clinic site.

[b] Variable was not included as a covariate because it did not add significantly to the model.

At baseline, higher SAT was associated with older age, being female, and lower 25[OH]D level in both populations. In addition, not being a current smoker, being married, and having a lower level of vigorous physical activity was significantly associated with higher SAT in Hispanics but not African-Americans. In Hispanics, the proportion of the variance explained by vitamin D level was 0.05, or 23% of the total variance explained by the model. In African-Americans, vitamin D explained 8.7% of the total variance explained by the model.

In both Hispanics and African-Americans, lower 25[OH]D was associated with a higher VAT at baseline, adjusting for age, sex, and in Hispanics, marital status. The proportion of variance explained by vitamin D level was 0.03 in both populations, which accounted for slightly more of the total variance in Hispanics (9.7%) compared with African-Americans (7.3%).

25[OH]D was not associated with baseline VSR in Hispanics and only marginally associated with VSR in African-Americans.

We examined the association between $1,25[OH]_2D$ and adiposity measures without and with adjustment for 25[OH]D because the two have been shown to be correlated (24). Lower $1,25[OH]_2D$ level was significantly associated with BMI in both the Hispanic and African-American populations at baseline, adjusting for age, gender, and smoking status (Table 3). This remained statistically significant after adjusting for 25[OH]D levels.

At baseline, higher SAT was significantly associated with lower $1,25[OH]_2D$ level after adjusting for age, gender, energy expenditure from vigorous activity, and current smoking status in the Hispanic population. This statistically significant association remained after adjusting for 25[OH]D levels. For African-Americans, lower $1,25[OH]_2D$ level was statistically significantly associated with higher SAT after adjusting for age, gender, and education status.

Higher baseline VAT was significantly associated with lower $1,25[OH]_2D$ levels in both Hispanic and African-Americans, adjusting for age and gender. However, after adjusting for baseline 25[OH]D level, this association was no longer statistically significant in either population.

$1,25[OH]_2D$ was not associated with VSR in Hispanics but was significantly associated with VSR in African-Americans, after adjusting for age and gender. This statistically significant association in African-Americans did not persist after adjustment for 25[OH]D levels.

TABLE 3. Regression coefficients (± SE) from multivariable models of plasma 1,25[OH]$_2$D levels on adiposity phenotypes at the baseline IRAS Family Study visit

	Adiposity phenotype							
	BMI Covariate coefficients ± SE for 1,25[OH]$_2$D *P* value		SAT Covariate coefficients ± SE for 1,25[OH]$_2$D *P* value		VAT Covariate coefficients ± SE for 1,25[OH]$_2$D *P* value		VSR Covariate coefficients ± SE for 1,25[OH]$_2$D *P* value	
Baseline covariate	Hispanics (n = 917)	African-Americans (n = 439)	Hispanics (n = 870)	African-Americans (n = 410)	Hispanics (n = 904)	African-Americans (n = 438)	Hispanics (n = 845)	African-Americans (n = 402)
Model 1: 1,25[OH]$_2$D	−0.002 ± 0.0004 $P < 0.0001^b$	−0.002 ± 0.0008 $P = 0.002^b$	−0.04 ± 0.009 $P < 0.0001^a$	−0.03 ± 0.02 $P = 0.07^a$	−0.02 ± 0.005 $P = 0.0005$	−0.02 ± 0.008 $P = 0.012$	0.0008 ± 0.0008 $P = 0.36^c$	−0.003 ± 0.001 $P = 0.02^c$
Model 2: 1,25[OH]$_2$D (adjusting for 25[OH]D level)	−0.001 ± 0.0004 $P = 0.02^b$	−0.002 ± 0.0008 $P = 0.04^b$	−0.03 ± 0.009 $P = 0.004^a$	−0.01 ± 0.02 $P = 0.49^a$	−0.01 ± 0.005 $P = 0.07$	−0.007 ± 0.008 $P = 0.39$	−0.0008 ± 0.002 $P = 0.38^c$	−0.003 ± 0.002 $P = 0.10^c$

All models are adjusted for age and gender (at baseline); and Hispanic models are also adjusted for clinic site.

[a] Also adjusted for energy expenditure from vigorous activity, current smoking status, and education status (at baseline).

[b] Also adjusted current smoking status (at baseline).

[c] Also adjusted for education status (at baseline).

Change in adiposity between baseline and follow-up

Baseline 25[OH]D levels were not associated with change in BMI, VAT, or SAT in Hispanics or African-Americans between the baseline and follow-up visit (mean 5.3 yr later), although a few of the associations approached significance (Table 4). We examined this question without and with adjustment for the corresponding baseline adiposity phenotype because change in adiposity may be, in part, associated with the level of adiposity with which one starts. 25[OH]D was significantly associated with change in VSR in Hispanics ($P = 0.03$) but not African-Americans. After adjusting for baseline VSR, 25[OH]D was no longer significantly associated with change in VSR in Hispanics ($P = 0.06$). We also examined the association of 1,25[OH]$_2$D levels with the change phenotypes but did not see any significant associations (data not shown).

Discussion

In the IRAS Family Study, the baseline mean 25[OH]D levels were 16.6 ng/ml in the Hispanic-Americans and 11.0 ng/ml in the African-Americans, which are comparable with a 2000–2004 National Health and Nutrition Examination Survey in which the mean 25[OH]D was 22.0 ng/ml and 22.0 ng/ml in Hispanic-Americans and 16.0 ng/ml and 17.2 ng/ml for African-Americans, in 20- to 49- and 50- to 69-yr-olds, respectively (31). African-Americans and Hispanics typically have lower levels of 25[OH]D than Caucasians (18, 31, 32) as a result of darker skin pigmentation, which decreases the amount of UVB radiation penetrating the skin to produce vitamin D. In our study population, we previously reported that Hispanics have higher levels of 25[OH]D than African-Americans; this is not necessarily true of 1,25[OH]$_2$D levels (24). Both ethnic groups also have increased prevalence of obesity in children, adolescents, and adults (33), compared with Caucasians. Despite the fact that these two ethnic groups make up the largest, and in the case of Hispanics most rapidly growing, minority populations in the United States, little research has been done to identify factors that might influence obesity in these populations.

Our results are consistent with previous studies reporting an inverse relationship between vitamin D levels and increased adiposity (3–19, 22). Whereas we did not have

TABLE 4. Regression coefficients (± SE) from multivariable models of baseline plasma 25[OH]D levels on change in adiposity phenotypes between baseline and follow-up IRAS Family Study visits

	Adiposity phenotype							
	Change in BMI Coefficient ± SE. for 25[OH]D *P* value		Change in SAT Coefficient ± SE. for 25[OH]D *P* value		Change in VAT Coefficient ± SE. for 25[OH]D *P* value		Change in VSR Coefficient ± SE. for 25[OH]D *P* value	
Baseline covariate	Hispanics (n = 657)	African-Americans (n = 289)	Hispanics (n = 657)	African-Americans (n = 284)	Hispanics (n = 657)	African-Americans (n = 284)	Hispanics (n = 646)	African-Americans (n = 284)
25[OH]D	0.01 ± 0.01 $P = 0.38$	−0.04 ± 0.03 $P = 0.15$	−0.25 ± 0.36 $P = 0.50$	−1.21 ± 0.85 $P = 0.16$	−0.27 ± 0.18 $P = 0.13$	−0.43 ± 0.35 $P = 0.22$	−0.001 ± 0.0005 $P = 0.06$	−0.0004 ± 0.0009 $P = 0.69$

All models are adjusted for age, age^2, gender, and the corresponding adiposity phenotype at baseline; models of Hispanics are also adjusted for clinic site.

a measure of total body fat (*i.e.* via dual-energy x-ray absorptiometry), previous studies have not shown a difference between BMI and total body fat measures in terms of the direction of association with vitamin D levels (11, 13–15, 19). Because anthropometric indicators of adiposity such as BMI are traditionally weaker than direct measures of adiposity, it is likely that we are underestimating the association between vitamin D and overall adiposity by using BMI as our estimate of total body fat. Only one other study (34) used CT-derived visceral fat or sc fat areas, which provide precise measures of type and depot of body fat, in 90 young Caucasian and Hispanic women. Similar to that study, we showed that lower 25[OH]D levels were associated with higher SAT, the sc fat found just below the skin's surface and an overall measure of adiposity, as well as higher VAT, the visceral fat found in the abdomen surrounding vital organs and a measure of central adiposity, suggesting that vitamin D levels are associated with both types of fat. Our study extends the previous study's (34) findings to men, older adults, and African-Americans.

It has been hypothesized that low 25[OH]D levels are associated with increased adiposity due to the decreased bioavailability of vitamin D that is sequestered in fat cells (16). Whereas our study cannot refute this, our observation that in African-Americans, decreased 25[OH]D is associated with higher VSR, a measure of relative distribution of fat rather than quantity of fat, suggests that the adiposity/25[OH]D relationship may not be as simple as increased sequestering of vitamin D and may indicate that greater amounts of visceral fat relative to sc fat could have an impact on vitamin D levels, or alternatively, that the greater relative amount of visceral fat and decreased vitamin D levels may be markers of the same condition. A recent *in vitro* study showed that vitamin D can inhibit leptin secretion, which may be a mechanism by which vitamin D can contribute to the maintenance of body mass (35). However, due to VSR's marginal association ($P = 0.049$) and that the association was not seen in Hispanic-Americans suggests that this needs to be confirmed in other populations. Our observation that higher BMI and SAT were associated with lower $1,25[OH]_2D$, after controlling for 25[OH]D levels, also suggests a more complex relationship between vitamin D and adiposity than simply attributing this association to vitamin D bioavailability, particularly in light of the recent observation that adipogenesis may be inhibited by $1,25[OH]_2D$ (21).

To further explore what role vitamin D plays in adiposity, we examined whether baseline vitamin D levels were associated with 5-yr change in adiposity. The lack of a significant association between baseline 25[OH]D and $1,25[OH]_2D$ levels and change in adiposity indicates that vitamin D levels are not a good predictor of increases (or decreases) of fat in the future. To our knowledge this is the first study to look at vitamin D levels as a predictor of change in adiposity in an observational setting. Whereas two vitamin D supplementation trials showed no significant difference in BMI changes between the placebo and intervention groups (36, 37), another noted a small effect on weight gain prevention in the intervention (vitamin D plus calcium) compared with placebo (38), indicating that vitamin D may play a role in weight loss. We measured vitamin D levels only at baseline in the IRAS Family Study participants. It would have been beneficial to have a second measure of vitamin D at follow-up to examine true longitudinal relationships between vitamin D and adiposity. Clearly there is a need for further vitamin D intervention studies of sufficient size to help clarify the nature of this association.

In summary, vitamin D levels (25[OH]D and $1,25[OH]_2D$) were inversely associated with baseline BMI, SAT, VAT, and VSR in Hispanic- and/or African-Americans but were not associated with 5-yr change in adiposity. Whereas it is clear that some portion of the inverse relationship between vitamin D and adiposity may be attributed to the sequestering of vitamin D in fat stores, observed associations between body fat distribution and 25[OH]D, and between $1,25[OH]_2D$ and adiposity suggest more complex relationships that should be explored to elucidate the overall role of vitamin D in health.

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