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Vitamin D Status Is Associated With Arterial Stiffness and Vascular Dysfunction in Healthy Humans

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Objectives The primary objective of this study was to elucidate mechanisms underlying the link between vitamin D status and cardiovascular disease by exploring the relationship between 25-hydroxyvitamin D (25-OH D), an established marker of vitamin D status, and vascular function in healthy adults.

Background Mechanisms underlying vitamin D deficiency-mediated increased risk of cardiovascular disease remain unknown. Vitamin D influences endothelial and smooth muscle cell function, mediates inflammation, and modulates the renin-angiotensin-aldosterone axis. We investigated the relationship between vitamin D status and vascular function in humans, with the hypothesis that vitamin D insufficiency will be associated with increased arterial stiffness and abnormal vascular function.

Methods We measured serum 25-OH D in 554 subjects. Endothelial function was assessed as brachial artery flow-mediated dilation, and microvascular function was assessed as digital reactive hyperemia index. Carotid-femoral pulse wave velocity and radial tonometry-derived central augmentation index and subendocardial viability ratio were measured to assess arterial stiffness.

Results Mean 25-OH D was 31.8 ± 14 ng/ml. After adjustment for age, sex, race, body mass index, total cholesterol, low-density lipoprotein, triglycerides, C-reactive protein, and medication use, 25-OH D remained independently associated with flow-mediated vasodilation ($\beta = 0.1$, $p = 0.03$), reactive hyperemia index ($\beta = 0.23$, $p < 0.001$), pulse wave velocity ($\beta = -0.09$, $p = 0.04$), augmentation index ($\beta = -0.11$, $p = 0.03$), and subendocardial viability ratio ($\beta = 0.18$, $p = 0.001$). In 42 subjects with vitamin D insufficiency, normalization of 25-OH D at 6 months was associated with increases in reactive hyperemia index (0.38 ± 0.14, $p = 0.009$) and subendocardial viability ratio (7.7 ± 3.1, $p = 0.04$), and a decrease in mean arterial pressure (4.6 ± 2.3 mm Hg, $p = 0.02$).

Conclusions Vitamin D insufficiency is associated with increased arterial stiffness and endothelial dysfunction in the conductance and resistance blood vessels in humans, irrespective of traditional risk burden. Our findings provide impetus for larger trials to assess the effects of vitamin D therapy in cardiovascular disease. (J Am Coll Cardiol 2011;58:186–92) © 2011 by the American College of Cardiology Foundation

Vitamin D deficiency is a highly prevalent condition affecting 30% to 50% of the U.S. population and is associated with hypertension, insulin resistance, and left ventricular hypertrophy (1,2). More recently, vitamin D deficiency has been implicated as a risk factor for cardiovascular disease and overall mortality in the general population (3,4). Although the link between the traditional role of vitamin D in bone and calcium metabolism has been extensively studied, the mechanisms by which vitamin D deficiency confers vascular risk remain unexplained.

Vitamin D receptors and 1-alpha-hydroxylase, which converts vitamin D into the hormonal 1,25-dihydroxyvitamin D form, are present in many tissues including endothelial cells (5). Endothelial cell vitamin D receptors are up-regulated under stress, and its binding with the hormonal vitamin D ligand modulates response elements in the vascular endothelial growth factor promoter (6). Vitamin D also affects

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the vascular wall by regulating the renin-angiotensin-aldosterone axis and exerts anti-proliferative effects on vascular smooth muscle (7). Moreover, vitamin D plays a crucial role in immune modulation by regulating lymphocyte and monocyte/macrophage differentiation and release of inflammatory cytokines. This in turn might determine monocyte infiltration and cholesterol retention in the vascular wall (8,9).

It is well established that vascular endothelial dysfunction and arterial stiffness precede and contribute to the development of cardiovascular disease and are both predictors of long-term morbidity and mortality (10,11). We investigated the mechanisms underlying the link between vitamin D insufficiency and cardiovascular disease by exploring the relationship between 25-hydroxyvitamin D (25-OH D), an established marker of vitamin D status, and vascular function. Our hypothesis was that vitamin D insufficiency will be associated with increased arterial stiffness and abnormal vascular function.

Methods

Subjects. A total of 554 healthy participants, 20 to 79 years of age, were recruited by advertisement or by invitation to employees of Emory University and Georgia Institute of Technology, as part of the Emory Predictive Health Initiative. Subjects were free of any acute illness and had normal performance status. Hypertension, hypercholesterolemia, and diabetes mellitus were defined according to the Seventh Report of the Joint National Committee on the Prevention, Detection, Evaluation, and Treatment of High Blood Pressure; Adult Treatment Panel III; and American Diabetes Association criteria, respectively (12–14). Vascular testing and blood draws were performed after an overnight fast. Ninety-one subjects with serum 25-OH D levels <30 ng/ml at baseline testing had repeat measurements performed after 6 months. The study was approved by the Emory University Institutional Review Committee. Informed consent was obtained from all subjects.

Laboratory tests. Blood samples were obtained for a lipid profile, and metabolic panel, urine albumin/creatinine ratio, high-sensitivity C-reactive protein (CRP), and serum 25-OH D levels (liquid chromatography and tandem mass spectrometry) were measured by commercially available assays (Quest Diagnostics, Madison, New Jersey). The detection limit for the 25-OH D was 4 ng/ml, with a previously reported inter-assay variation coefficient of approximately 10% (15).

Arterial stiffness testing. After an initial rest period of 10 min with subjects in a supine position in a quiet, temperature-controlled room, blood pressure was measured 3 times at 5-min intervals by an automatic device (Omron, Kyoto, Japan) from the dominant arm and documented as the mean value of the final 2 measurements.

Pulse wave velocity (PWV), augmentation index (AIX), and subendocardial viability ratio (SEVR) were derived with the Sphygmocor device (Atcor Medical, Sydney, Australia), as previously described (16). Briefly, the setup consists of a handheld tonometer attached to a device and a laptop computer. High-fidelity sequential pressure waveforms are obtained by placing the tonometer on the radial artery of the dominant wrist. The device then applies a transfer function to these peripheral measurements to estimate central (aortic) pressure parameters and the degree of pressure augmentation secondary to reflected waves from the periphery. This permits derivation of an AIX (augmented pressure/total central pulse pressure) and SEVR (area under diastolic phase/systolic phase), which are considered complex and composite markers of wave reflections and arterial stiffening. Due to its sensitivity to heart rate, a standardized value to 75 beats/min is calculated and used for AIX for the purpose of this study.

Abbreviations and Acronyms

25-OH D = 25-hydroxyvitamin D
AIX = augmentation index
CRP = C-reactive protein
FMD = (brachial artery) flow-mediated dilation
PWV = pulse wave velocity
RHI = reactive hyperemia index
SEVR = subendocardial viability ratio

The PWV was determined by acquiring waveforms at the carotid and femoral arterial sites with electrocardiogram gating. Velocity (distance/time in m/s) was calculated by measuring the time interval between electrocardiogram R-wave and the recorded waveforms at each site, whereas distance between sites was measured manually. Quality control indexes were evaluated at the time of study and nonacceptable readings discarded and repeated. Reproducibility studies in our laboratory on 9 subjects on consecutive days have demonstrated a coefficient of variation of 3.8%, 13.8%, and 20.3% for PWV, SEVR, and AIX, respectively.

Flow-mediated dilation. Endothelium-dependent brachial artery flow-mediated dilation (FMD) was measured to evaluate endothelium-dependent vascular function. The detailed methodology for performing this test has been described previously (17). In our laboratory, the mean difference in FMD between assessments performed in 11 subjects on consecutive days was 1.26 ± 0.76%, with a correlation coefficient of 0.75. The mean difference in the FMD between 2 readings of the same 11 measurements was 0.82 ± 0.48% (r = 0.97).

Pulsatile arterial tonometry. During the FMD testing described in the preceding text, digital vascular function was also assessed simultaneously as fingertip reactive hyperemia (Endo-PAT2000, Itamar Medical, Caesarea, Israel) as described previously (18). Briefly, pneumatic plethysmographs apply uniform pressure at both index fingers that allows for measurement of minute, pulsatile volume changes, generating a pulse wave tracing. Reactive hyperemia index (RHI) is defined as the ratio of post-deflation to baseline pulse amplitude in the hyperemic finger divided by that in the contra-lateral finger.

Statistical considerations. Study variables are described as the mean ± SD (unless otherwise specified) for continuous variables or as counts or proportions for categorical variables. Age, body mass index, 25-OH D level, CRP, lipid profile, FMD, RHI, PWV, AIX, and SEVR values were treated as continuous variables. Diagnosed hypertension, hypercholesterolemia, diabetes mellitus, smoking, sex, medication use (anti-hypertensive drugs, lipid or glucose lowering agents), and vitamin D supplementation (≥10 μg/day) were categorical variables. Continuous variables were tested for normality with the Kolmogorov-Smirnov criterion. Skewed variables were log transformed and tested again for normality before any parametric analysis.

Univariate correlations between serum 25-OH D concentrations and measured parameters were performed with Pearson's correlation. Multivariate linear regression models were constructed to determine relationships between vitamin D status and vascular function parameters before and after adjustment for age, sex, race, body mass index, total cholesterol, low-density lipoprotein, triglycerides, CRP, and medication use. Group differences were evaluated by Student *t* tests or 1-way analysis of variance.

For follow-up analysis, participants with vitamin D insufficiency at baseline (n = 91) were divided into those who normalized their 25-OH D ≥30 ng/ml levels and those who remained insufficient (25-OH <30 ng/ml) after 6 months. We compared average changes in mean arterial pressure, FMD, RHI, PWV, AIX, and SEVR (baseline − 6-month value) between groups with unpaired samples *t* tests. Paired samples *t* tests were performed for comparison of within-group differences. Data are presented as mean ± SD in the text and as specified in the legends of Figures 1, 2, and 3. Statistical significance was based on 2-tailed tests, and p values ≤0.05 were considered significant. Analyses were performed with SPSS (version 17.0, SPSS, Inc., Chicago, Illinois).

Results

Baseline characteristics of the 554 subjects are shown in Table 1. Mean 25-OH D level was 31.8 ng/ml (range 4 to 96 ng/ml), and 47% of subjects had insufficient 25-OH D levels (<30 ng/ml).

Relationship between vitamin D status and subject characteristics. Serum 25-OH D correlated with body mass index ($r = -0.23$, $p < 0.001$) and high-density lipoprotein levels ($r = 0.2$, $p < 0.001$). Among the categorical variables, 25-OH D was significantly lower in women, African Americans, Hispanics, and those with hypertension or diabetes (Table 2). Subjects taking regular vitamin D supplementation had higher 25-OH D levels (38 ng/ml vs.

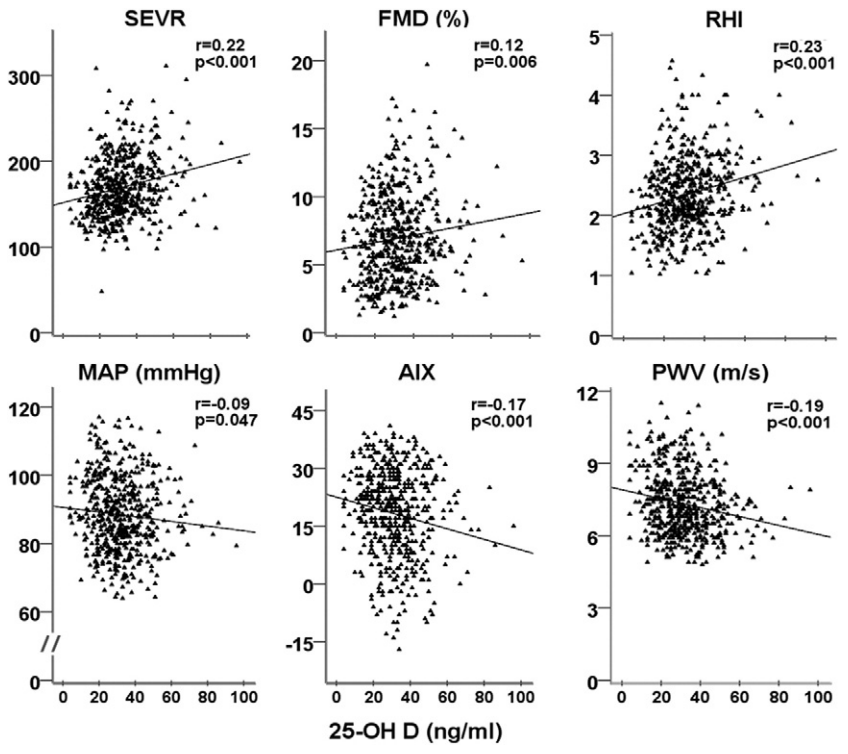

Figure 1 Relationship Between Serum 25-OH D, Arterial Stiffness, and Vascular Function

25-OH D = 25-hydroxyvitamin D (ng/ml); AIX = augmentation index; FMD = brachial artery flow mediated dilation (%); MAP = mean arterial pressure (mm Hg); PWV = pulse wave velocity (m/s); RHI = reactive hyperemia index; SEVR = subendocardial viability ratio. p values are for 2-tailed test of significance.

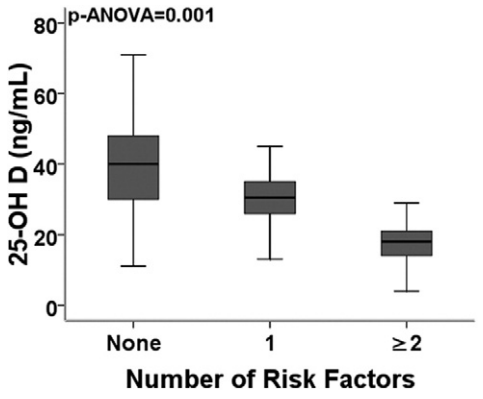

Figure 2 **Cardiovascular Risk Burden and Levels of Serum 25-OH D**

Risk factors included: age (men older than 55 or women older than 65 years), presence of diabetes mellitus, hypertension, hypercholesterolemia, or smoking history. Box plots: the **middle band** represents median, **bottom and top of the box** represent lower and upper quartiles, and **end of whiskers** represent highest and lowest values that are not outliers. p value for 1-way analysis of variance. n = 266, 128, and 150 for groups with 0, 1, or >2 risk factors, respectively. 25-OH D = 25-hydroxyvitamin D.

29 ng/ml, $p < 0.001$) and were significantly older (54 years vs. 46 years, $p = 0.001$). When the 544 subjects were divided on the basis of risk factor burden into groups with none, 1, or 2 or more risk factors, a stepwise decline in mean 25-OH D levels was observed with increasing risk factor burden (p analysis of variance = 0.012) (Fig. 2).

Relationship between vitamin D status and arterial stiffness. Lower levels of 25-OH D were associated with abnormalities in the indexes of arterial stiffness with higher AIX and PWV and lower SEVR (Fig. 1). After multivariate adjustment for age, sex, race, body mass index, total cholesterol, low-density lipoprotein, triglycerides, C-reactive protein, and medication use, 25-OH D remained independently associated with PWV (adjusted $R^2 = 0.32$, $\beta = -0.09$, $p = 0.04$), AIX (adjusted $R^2 = 0.42$, $\beta = -0.11$, $p = 0.03$), and SEVR (adjusted $R^2 = 0.16$, $\beta = 0.18$, $p = 0.001$).

Relationship between vitamin D status and vascular function. Lower levels of 25-OH D were associated with abnormalities in the indexes of vascular function with lower FMD and RHI (Fig. 1). Moreover, 25-OH D levels had an independent association with FMD (adjusted $R^2 = 0.11$, $\beta = 0.1$, $p = 0.03$) and RHI (adjusted $R^2 = 0.09$, $\beta = 0.23$, $p < 0.001$) in multivariate analysis adjusting for the aforementioned variables.

In age-, sex-, and race-adjusted analyses, no significant correlation existed between log CRP and 25-OH D.

Subgroup analysis in subjects free of cardiovascular risk factors. A secondary analysis was performed in a subgroup of 233 "healthy" subjects with absence of traditional cardiovascular risk factors, to explore the association between levels of 25-OH D and vascular function independent of the confounding effects of risk factors and vitamin D intake. After multivariate adjustment for age, sex, race, body mass index, systolic arterial pressure, and total cholesterol/high-density lipoprotein ratio, 25-OH D levels remained inde-

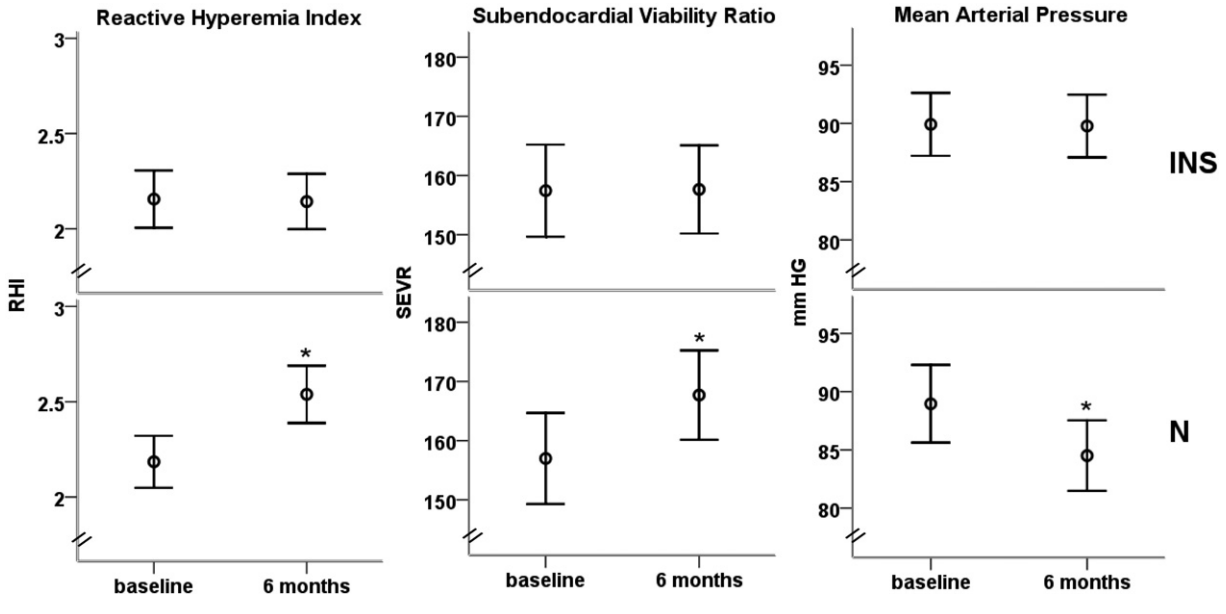

Figure 3 **Follow-Up Measurements in Subjects With Vitamin D Insufficiency**

Mean arterial pressure, reactive hyperemia index, and subendocardial viability ratio at baseline and after 6 months in 49 subjects who remained insufficient (25-OH D <30 ng/ml) after 6 months **(top panels)**, and those in whom vitamin D levels normalized after 6 months (n = 42, **bottom panels**). **Error bars** represent standard error. *Significant between and within-group differences.

JACC Vol. 58, No. 2, 2011
July 5, 2011:186–92

Table 1 Demographic Characteristics

Age (yrs)	47 ± 13
Men	45
Caucasian	71
African American	22
Hispanic	2
Body mass index (kg/m^2)	26 ± 5
Diabetes	5
Hypertension	19
Hypercholesterolemia	25
Smoking	3
25-hydroxyvitamin D (ng/ml)	32 ± 14
Calcium (mg/dl)	9.4 ± 0.3
Cholesterol/high-density lipoprotein	3.1 ± 0.9
Triglycerides (mg/dl)	94 ± 46
hs-CRP (mg/dl)	0.6 ± 0.8
Statin use	25
Vitamin D supplements	32
Framingham Risk Score	2.7 ± 6.2

Values are mean ± SD or %.
CRP = C-reactive protein.

pendently associated with AIX (adjusted $R^2 = 0.44$, $\beta = -0.02$, $p < 0.001$), PWV (adjusted $R^2 = 0.2$, $\beta = 0.02$, $p = 0.005$), FMD (adjusted $R^2 = 0.12$, $\beta = -0.01$, $p = 0.04$), and RHI (adjusted $R^2 = 0.07$, $\beta = 0.02$, $p = 0.003$) in this cohort but accounted for small changes in variability.

Follow-up. Of the 91 participants with vitamin D insufficiency, 42 had normalized vitamin D status (25-OH D ≥30 ng/ml) and 49 subjects remained in insufficient status after 6 months. Subjects who normalized their 25-OH D levels had significant increases in RHI (0.38 ± 0.29, p = 0.009) and SEVR (7.7 ± 11.4, p = 0.04) and a decrease in mean arterial pressure (4.6 ± 5.6 mm Hg, p = 0.02). These differences were significantly greater when compared with subjects where 25-OH D levels remained in the insufficient range (0.38 ± 0.29 vs. −0.07 ± 0.33, p = 0.01; 7.7 ± 11.4 vs. −1.1 ± 9.5, p = 0.03; −4.6 ± 5.6 mm Hg vs. −0.9 ± 4.7 mm Hg, p = 0.02, for RHI, SEVR, and mean arterial pressure, respectively) (Fig. 3). Thus, subjects with corrected vitamin D insufficiency had improved microvascular vasodilation and arterial stiffness.

Discussion

In a study that comprehensively assesses vascular function in a community-based asymptomatic population, we demonstrated an association between vitamin D status and several indexes of vascular health, including arterial stiffness and endothelial function. We studied a large multiethnic population over a wide age range but with a relatively low risk factor burden, and we employed multiple established modalities to assess subclinical vascular dysfunction. Increased arterial stiffness—assessed by measurements of PWV, AIX, and SEVR—was associated with lower 25-OH D levels. In addition, lower 25-OH D levels were associated with worse vascular endothelial function, assessed in the conductance vessels with FMD and in the microvasculature with pulsatile digital arterial tonometry. Moreover, subgroup analysis performed in selected individuals free of all vascular risk factors in order to exclude the confounding effects of risk factors on vascular function also revealed an independent association between vitamin D status and vascular function. These observations are consistent with the well-established associations between vitamin D deficiency and a broad range of cardiovascular disorders and risk factors (1,3).

25-hydroxyvitamin D (25-OH D) is considered to be the best indicator of vitamin D status in those with normal kidney function and reflects the level of circulating substrate for the tightly regulated hydroxylation into the active, hormonal form of vitamin D (1,25-OH_2 D) (1). Alternatively, measurement of 1,25-OH_2 D is performed in chronic kidney disease, as decreases in renal 1-α-hydroxylase activity often result in low 1,25-OH_2 D levels and hypocalcemia.

Previous small studies have also shown increased AIX and lower FMD in hemodialysis patients with lower levels of 1,25-OH_2 D, but our findings in a community-based population are novel (19,20). In hypertensive patients, levels of 25-OH D were inversely associated with plethysmography-derived calf vascular resistance (21), and the third National Health and Nutrition Examination Survey found increased pulse pressure, a nonspecific marker of arterial compliance, with vitamin D deficiency (22). Finally, few well-designed interventional trials have explored whether vascular function can be improved by vitamin D supplementation (23,24).

In experimental studies vitamin D modulates endothelial cell function by decreasing expression of adhesion molecules, providing protection against advanced glycation products, by reduction in endothelium-dependent contractions and modulation of calcium influx (25–27). Vitamin D

Table 2 Relationship Between Vitamin D Status and Subject Characteristics

	ng/ml	p Value
Sex		
Men	34 ± 14	0.03
Women	31 ± 13	
Race		
Caucasian	35 ± 14	<0.001
African American	23 ± 12	
Diabetes		
Yes	22 ± 8	0.001
No	32 ± 14	
Hypertension		
Yes	26 ± 15	0.006
No	34 ± 13	
Hyperlipidemia		
Yes	32 ± 14	NS
No	32 ± 14	
Vitamin D supplement		
Yes	38 ± 12	<0.001
No	29 ± 14	

Values are mean ± SD serum 25-hydroxyvitamin D concentration. p values were derived from the Student independent samples *t* test.

deficiency also activates the renin-angiotensin-aldosterone system, causes proliferation of vascular smooth muscle cells, activates macrophage invasion of the vascular wall, promotes calcification, and increases parathyroid hormone release, effects that are ameliorated by vitamin D supplementation (7,25,28). Finally, vitamin D might modulate adaptive immunity by reducing inflammatory cytokine gene expression, alternating circulating subsets of T-cells, and reducing inflammation (29,30). In clinical studies, an increased incidence of adverse cardiovascular events has been found in the Framingham cohort with decreased serum 25-OH D, and community-based studies have observed associations between vitamin D deficiency and cardiovascular disease and its risk factors, as in our population (3,4).

Our findings of increased arterial stiffness and endothelial dysfunction with low 25-OH D provide mechanistic explanation of how depressed vitamin D status, by precipitating vascular dysfunction, might predispose individuals to a higher risk for the development of cardiovascular disease and adverse events. Brachial artery FMD, a measure of endothelial function and nitric oxide bioavailability, is also an independent predictor of future adverse cardiovascular events (31). Persistence of endothelial dysfunction despite therapeutic interventions is associated with worse prognosis, and normalization of FMD leads to improved outcomes (32). Similarly, RHI has been negatively associated with cardiovascular risk factors, and low RHI seems to be associated with worse long-term outcome (33). Similarly, arterial stiffness indexes including AIX and SEVR are independent predictors of adverse outcomes (16,34).

Increased arterial stiffness and endothelial dysfunction are also strong predictors for future development of hypertension (35). In this context, the majority of large cross-sectional studies have demonstrated an inverse relationship between 25-OH D levels and blood pressure, and the risk of future hypertension seems to be greater in those with vitamin D deficiency (36,37). Although interventional trials with vitamin D supplementation in hypertension are sparse, a study in post-menopausal women reported a 7.8-mm Hg reduction in systolic blood pressure with vitamin D replacement (38).

Our findings of higher serum 25-OH D in men compared with women, Caucasians compared with African Americans and Hispanics, and in participants taking vitamin D have been previously reported (39). Similarly, higher body mass index, lower high-density lipoprotein, and presence of diabetes or hypertension were associated with decreased 25-OH D levels (1) (Table 2).

It is noteworthy that our subjects were asymptomatic and had normal serum calcium and urinary albumin/creatinine ratio (Table 1). In addition, all subjects had an estimated glomerular filtration rate >60 ml/min/1.73 m^2, and none reported use of high-dose vitamin D supplements (>2,000 IU daily) at baseline. Although the highest 25-OH D level was 96 ng/ml in our population, levels >240 ng/ml have been associated with displacement of bound 1,25-OH_2 D and subsequent hypercalcemia (40). Although vitamin D toxicity can be serious, vitamin D supplementation normalizes 25-OH D levels and exerts antirachitic effects. Finally, novel vitamin D analogs with less calciphylactic effects need to be investigated in cardiovascular disease.

Study limitations. The cross-sectional, observational design of our current study precludes definitive conclusions regarding the causal relationship between vascular dysfunction and hypovitaminosis D. Also, the magnitude of contribution of hypovitaminosis D to vascular dysfunction might be variable and modest. However, the improvements we observed in some of these subclinical markers of vascular health with normalization of vitamin D insufficiency help confirm the link between vitamin D status and vascular health and also highlight a potential role for vitamin D therapy in reducing cardiovascular risk. We did not find significant improvements in all measures of vascular dysfunction in the follow-up study in those who normalized vitamin D levels. This might be because we were underpowered to observe such a change, the study was observational, and the supplementation was variable. However, vitamin D supplementation seems to significantly improve vascular function (23,24).

It is also possible that other subject characteristics such as physical activity might account for the associations we observed. Increased physical activity is known to improve vascular function and might also improve vitamin D status by increasing sunlight exposure. We previously demonstrated an independent correlation between vitamin D status and cardiovascular fitness, measured by cardiopulmonary exercise testing (41).

Conclusions

Vitamin D insufficiency is associated with increased arterial stiffness and endothelial dysfunction in the conductance and resistance blood vessels in humans, irrespective of traditional risk factor burden. Normalization of vitamin D status in insufficient individuals is associated with improvements in several parameters of vascular function. Our findings provide impetus for larger trials to assess the effect of vitamin D therapy on prevention of cardiovascular disease in individuals with vitamin D insufficiency.

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REFERENCES

1. Holick MF. Vitamin D deficiency. N Engl J Med 2007;357:266–81.
2. Judd SE, Nanes MS, Ziegler TR, Wilson PW, Tangpricha V. Optimal vitamin D status attenuates the age-associated increase in systolic blood pressure in white Americans: results from the third National Health and Nutrition Examination Survey. Am J Clin Nutr 2008;87:136–41.
3. Wang TJ, Pencina MJ, Booth SL, et al. Vitamin D deficiency and risk of cardiovascular disease. Circulation 2008;117:503–11.

4. Melamed ML, Michos ED, Post W, Astor B. 25-hydroxyvitamin D levels and the risk of mortality in the general population. Arch Intern Med 2008;168:1629–37.
5. Zehnder D, Bland R, Chana RS, et al. Synthesis of 1,25-dihydroxyvitamin D(3) by human endothelial cells is regulated by inflammatory cytokines: a novel autocrine determinant of vascular cell adhesion. J Am Soc Nephrol 2002;13:621–9.
6. Raymond MA, Desormeaux A, Labelle A, et al. Endothelial stress induces the release of vitamin D-binding protein, a novel growth factor. Biochem Biophys Res Commun 2005;338:1374–82.
7. Li YC, Kong J, Wei M, Chen ZF, Liu SQ, Cao LP. 1,25-Dihydroxyvitamin D(3) is a negative endocrine regulator of the renin-angiotensin system. J Clin Invest 2002;110:229–38.
8. Kreindler JL, Steele C, Nguyen N, et al. Vitamin D3 attenuates Th2 responses to Aspergillus fumigatus mounted by CD4+ T cells from cystic fibrosis patients with allergic bronchopulmonary aspergillosis. J Clin Invest 2010;120:3242–54.
9. Oh J, Weng S, Felton SK, et al. 1,25(OH)2 vitamin d inhibits foam cell formation and suppresses macrophage cholesterol uptake in patients with type 2 diabetes mellitus. Circulation 2009;120:687–98.
10. Quyyumi AA. Prognostic value of endothelial function. Am J Cardiol 2003;91:19H–24H.
11. Weber T, Auer J, O'Rourke MF, et al. Arterial stiffness, wave reflections, and the risk of coronary artery disease. Circulation 2004; 109:184–9.
12. Lenfant C, Chobanian AV, Jones DW, Roccella EJ; Joint National Committee on the Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. Seventh Report of the Joint National Committee on the Prevention, Detection, Evaluation, and Treatment of High Blood Pressure (JNC 7): resetting the hypertension sails. Hypertension 2003;41:1178–9.
13. Executive Summary of the Third Report of The National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). JAMA 2001;285:2486–97.
14. Genuth S, Alberti KG, Bennett P, et al. Follow-up report on the diagnosis of diabetes mellitus. Diabetes Care 2003;26:3160–7.
15. Holick MF, Biancuzzo RM, Chen TC, et al. Vitamin D2 is as effective as vitamin D3 in maintaining circulating concentrations of 25-hydroxyvitamin D. J Clin Endocrinol Metab 2008;93:677–81.
16. Laurent S, Cockcroft J, Van Bortel L, et al. Expert consensus document on arterial stiffness: methodological issues and clinical applications. Eur Heart J 2006;27:2588–605.
17. Corretti MC, Anderson TJ, Benjamin EJ, et al. Guidelines for the ultrasound assessment of endothelial-dependent flow-mediated vasodilation of the brachial artery: a report of the International Brachial Artery Reactivity Task Force. J Am Coll Cardiol 2002;39:257–65.
18. Hamburg NM, Keyes MJ, Larson MG, et al. Cross-sectional relations of digital vascular function to cardiovascular risk factors in the Framingham Heart Study. Circulation 2008;117:2467–74.
19. Andrade J, Er L, Ignaszewski A, Levin A. Exploration of association of 1,25-OH2D3 with augmentation index, a composite measure of arterial stiffness. Clin J Am Soc Nephrol 2008;3:1800–6.
20. London GM, Guerin AP, Verbeke FH, et al. Mineral metabolism and arterial functions in end-stage renal disease: potential role of 25-hydroxyvitamin D deficiency. J Am Soc Nephrol 2007;18:613–20.
21. Duprez D, de Buyzere M, de Backer T, Clement D. Relationship between vitamin D3 and the peripheral circulation in moderate arterial primary hypertension. Blood Press 1994;3:389–93.
22. Martins D, Wolf M, Pan D, et al. Prevalence of cardiovascular risk factors and the serum levels of 25-hydroxyvitamin D in the United States: data from the Third National Health and Nutrition Examination Survey. Arch Intern Med 2007;167:1159–65.
23. Sugden JA, Davies JI, Witham MD, Morris AD, Struthers AD. Vitamin D improves endothelial function in patients with Type 2 diabetes mellitus and low vitamin D levels. Diabet Med 2008;25: 320–5.
24. Tarcin O, Yavuz DG, Ozben B, et al. Effect of vitamin D deficiency and replacement on endothelial function in asymptomatic subjects. J Clin Endocrinol Metab 2009;94:4023–30.
25. Martinesi M, Bruni S, Stio M, Treves C. 1,25-Dihydroxyvitamin D3 inhibits tumor necrosis factor-alpha-induced adhesion molecule expression in endothelial cells. Cell Biol Int 2006;30:365–75.
26. Talmor Y, Golan E, Benchetrit S, et al. Calcitriol blunts the deleterious impact of advanced glycation end products on endothelial cells. Am J Physiol Renal Physiol 2008;294:F1059–64.
27. Wong MS, Delansorne R, Man RY, Vanhoutte PM. Vitamin D derivatives acutely reduce endothelium-dependent contractions in the aorta of the spontaneously hypertensive rat. Am J Physiol Heart Circ Physiol 2008;295:H289–96.
28. Helming L, Bose J, Ehrchen J, et al. 1alpha,25-Dihydroxyvitamin D3 is a potent suppressor of interferon gamma-mediated macrophage activation. Blood 2005;106:4351–8.
29. Yu S, Bruce D, Froicu M, Weaver V, Cantorna MT. Failure of T cell homing, reduced CD4/CD8alphaalpha intraepithelial lymphocytes, and inflammation in the gut of vitamin D receptor KO mice. Proc Natl Acad Sci U S A 2008;105:20834–9.
30. Pittas AG, Lau J, Hu FB, Dawson-Hughes B. The role of vitamin D and calcium in type 2 diabetes. A systematic review and meta-analysis. J Clin Endocrinol Metab 2007;92:2017–29.
31. Benjamin EJ, Larson MG, Keyes MJ, et al. Clinical correlates and heritability of flow-mediated dilation in the community: the Framingham Heart Study. Circulation 2004;109:613–9.
32. Kitta Y, Obata JE, Nakamura T, et al. Persistent impairment of endothelial vasomotor function has a negative impact on outcome in patients with coronary artery disease. J Am Coll Cardiol 2009;53:323–30.
33. Rubinshtein R, Kuvin JT, Soffler M, et al. Assessment of endothelial function by non-invasive peripheral arterial tonometry predicts late cardiovascular adverse events. Eur Heart J 2010;31:1142–8.
34. Sutton-Tyrrell K, Najjar SS, Boudreau RM, et al. Elevated aortic pulse wave velocity, a marker of arterial stiffness, predicts cardiovascular events in well-functioning older adults. Circulation 2005; 111:3384–90.
35. Quyyumi AA, Patel RS. Endothelial dysfunction and hypertension: cause or effect? Hypertension 2010;55:1092–4.
36. Pilz S, Tomaschitz A, Ritz E, Pieber TR. Vitamin D status and arterial hypertension: a systematic review. Nat Rev Cardiol 2009;6: 621–30.
37. Forman JP, Giovannucci E, Holmes MD, et al. Plasma 25-hydroxyvitamin D levels and risk of incident hypertension. Hypertension 2007;49:1063–9.
38. Pfeifer M, Begerow B, Minne HW, Nachtigall D, Hansen C. Effects of a short-term vitamin D(3) and calcium supplementation on blood pressure and parathyroid hormone levels in elderly women. J Clin Endocrinol Metab 2001;86:1633–7.
39. Scragg R, Sowers M, Bell C. Serum 25-hydroxyvitamin D, ethnicity, and blood pressure in the Third National Health and Nutrition Examination Survey. Am J Hypertens 2007;20:713–9.
40. Vieth R. Vitamin D toxicity, policy, and science. J Bone Miner Res 2007;22 Suppl 2:V64–8.
41. Al Mheid, Ronnie Ramadan, Nino Kavtaradze, et al. Vitamin D levels are associated with exercise capacity and measures of endothelial function in healthy humans (abstr). Circulation 2009;120:s551.

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