Vitamin D₃ supplementation (4000 IU/d for 1 y) eliminates differences in circulating 25-hydroxyvitamin D between African American and white men¹⁻⁴

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ABSTRACT
Background: African Americans suffer disproportionately from diabetes and cardiovascular disease and are significantly more likely to have suboptimal concentrations of circulating 25-hydroxyvitamin D [25(OH)D]. The results of epidemiologic and observational studies suggest that there is a link between vitamin D deficiency and the risk of cardiometabolic disorders, which underscores the importance of maintaining healthy concentrations of 25(OH)D.

Objective: The objective was to investigate whether daily supplementation with 4000 IU vitamin D₃ for 1 y would eliminate any disparities in circulating concentrations of 25(OH)D between African American and white men.

Design: Serum concentrations of 25(OH)D were measured every 2 mo in 47 subjects who received a daily oral dose of 4000 IU vitamin D₃ for 1 y.

Results: More than 90% of African Americans had serum concentrations of 25(OH)D <32 ng/mL, and approximately two-thirds had serum concentrations <20 ng/mL. Furthermore, there were significant disparities in serum concentrations of 25(OH)D between African American and white men. Supplementation with 4000 IU/d for 1 y eliminated any significant differences in circulating concentrations of 25(OH)D between African American and white men.

Conclusion: The results of this clinical study show the feasibility and efficacy of this approach in the elimination of hypovitaminosis D, which is a widespread health disparity among African Americans. This trial was registered at clinicaltrials.gov as NCT01045109.

INTRODUCTION
The exposure of skin to sunlight in the UVB range of the spectrum (290–315 nm) results in the photolytic conversion of 7-dehydrocholesterol to previtamin D₃, which is transformed into vitamin D₃ by thermally induced isomerization (1, 2). Vitamin D₃ can be obtained from the diet; however, it is distributed very poorly in natural foodstuffs. Increased skin pigmentation limits one’s ability to produce vitamin D₃ (3, 4). According to findings from NHANES 2001–2004, >90% of African Americans have serum concentrations of 25-hydroxyvitamin D [25(OH)D] <30 ng/mL (5). Furthermore, urban African Americans are 1.7 times more likely than their rural counterparts to exhibit hypovitaminosis D (6).

The results of retrospective and interventional studies suggest that to maximize skeletal integrity, circulating 25(OH)D should exceed 80 nmol (32 ng/mL) (7, 8). Some of these data, as well as additional studies, have been summarized in a recent review on the optimization of circulating 25(OH)D concentrations (9), which suggests that the current reference ranges for circulating 25(OH)D are too low; the desirable range of values for serum 25(OH)D in a sun-rich environment is 54–90 ng/mL (10, 11).

The recent emphasis on the noncalcemic functions of vitamin D concerns the realization that vitamin D deficiency has major implications for human health (12). There is an emerging scientific literature linking impaired vitamin D status with chronic conditions such as diabetes and cardiovascular disease. According to a systematic review of the current literature that screened >6000 references on vitamin D and cardiometabolic disorders (13), higher concentrations of circulating vitamin D among middle-age and elderly populations are associated with a substantial decrease in cardiovascular disease, type 2 diabetes, and metabolic syndrome. This association is particularly important for African Americans, who are affected by these pathologies in disproportionate numbers (14, 15). Furthermore, African Americans are significantly more likely to have suboptimal concentrations of circulating 25(OH)D (5).

Several studies suggest that intakes of 1000 IU/d increase serum 25(OH)D values to only slightly above 24 ng/mL (16–18). Vieth et al (19) examined the efficacy and safety of relatively high intakes of vitamin D₃ by assessing the effects of 1000 and 4000 IU/d in 61 adults for ≤5 mo. They found that vitamin D₃ at a dose of 4000 IU/d was effective in elevating the serum 25(OH)D concentration to ≥40 ng/mL. Until recently, higher-dose...
vitamin D₃ oral supplementation was not viewed as a viable treatment modality because of concerns about potential toxicity. This interventional study was aimed at investigating vitamin D₃ supplementation and its impact on hypovitaminosis D in African American men.

SUBJECTS AND METHODS

Study population

Eligible subjects were enrolled through the Clinical and Translational Research Center at the Medical University of South Carolina. Fifty-two male subjects diagnosed with early-stage, low-risk prostate cancer received vitamin D₃ supplementation at 4000 IU/d for 1 y. Their serum values of 25(OH)D were measured at enrollment and every 2 mo afterward for a total of 7 measurements per enrolled subject. Circulating concentrations of vitamin D [25(OH)D] were measured by radioimmunoassay as previously described (20, 21). BMI was calculated by using the formula weight in kilograms/(height in meters)². Information on BMI and 25(OH)D was collected every 2 mo. Forty-seven subjects completed 1 y of supplementation and were included in the analysis. Every 2 mo, we assessed the basic metabolic panel in all subjects enrolled in this interventional study, including calcium and phosphorus, and we measured the urinary calcium:creatinine ratio (in mg/mL:mg/mL, with normal values 0.22 in adult males) to monitor any potential toxicities from vitamin D₃ supplementation. The vitamin D₃ (cholecalciferol) administered for this study was manufactured by JR Carlson Laboratories Inc. This study was conducted under investigational new drug 77,839 (clinicaltrials.gov identifier: NCT01045109) and approved by the institutional review board of the Medical University of South Carolina.

Statistical analyses

Statistical analyses were performed in R version 2.14.2 (22) and in WinBugs version 1.4.3 (23). To be conservative, variance was assumed to be unequal, and Welch’s 2-sample t tests were used to compare mean age, BMI, and baseline 25(OH)D concentrations across races. Modeling of 25(OH)D over time was performed according to the model proposed by Heaney et al (24), who assumed that there are 2 parameters to define the relation between 25(OH)D concentrations and time since supplementation. Specifically, the exponential model was defined as follows:

\[
C(t) = C(0) + a(1 - e^{-kt})
\]

where \(t\) is time, \(C(t)\) is the 25(OH)D concentrations at time \(t\) [implying that \(C(0)\) is the baseline concentration], \(a\) is the increment to equilibrium produced by a given constant input, and \(k\) is the rate constant representing the proportion of total mass of 25(OH)D metabolized per time unit. This model was generalized to allow for separate \(a\) and \(k\) parameters for African American and white subjects. The model was estimated in WinBugs, which uses a Bayesian modeling framework and uses Gibbs sampling for generating posterior distributions of parameters. It was assumed that vitamin D concentrations at each time for each individual were normally distributed with mean \(C(t)\) and variance \(\sigma^2\). Prior distributions for parameters were as follows: \(\sigma\) and \(k\) were assumed to follow diffuse inverse gamma distributions, and \(a\) was assumed to follow a diffuse uniform distribution (with limits of 0–100). Five thousand iterations were performed and discarded, followed by 25,000 iterations, every fifth iteration of which was saved. Posterior distributions were based on the 5000 saved iterations. Medians of the posterior distributions served as our point estimates with 95% posterior intervals included to provide precision of estimates. Comparisons of parameters across races are based on the posterior probability estimates of the differences in parameters. Convergence was checked by rerunning the model with different starting values and comparing the resulting posterior distributions. The distribution of residuals was checked to ensure that our assumption of normality of residuals was appropriate. We also used longitudinal linear regression modeling with 3 df for time (ie, we included a linear, quadratic, and cubic term) to more flexibly model the relation between time and 25(OH)D to ensure that the exponential model was appropriate (and not too stringent) for our observed data. The 2 models provided very similar fits, which validates the appropriateness of the exponential model in this setting. Serum values of 25(OH)D were measured in nanograms per milliliter (ng/mL) and expressed as means ± SDs, as appropriate.

RESULTS

Subjects enrolled into the study were followed for 12 mo, and information on BMI and 25(OH)D was collected every 2 mo. Comparisons of age, BMI, and 25(OH)D at baseline and at study exit (12 mo) are provided in Table 1, which shows that African American and white subjects had comparable ages, baseline and in WinBugs version 1.4.3 (23). To be conservative, variance was assumed to be unequal, and Welch’s 2-sample t tests were used to compare mean age, BMI, and baseline 25(OH)D concentrations across races. Modeling of 25(OH)D over time was performed according to the model proposed by Heaney et al (24), who assumed that there are 2 parameters to define the relation between 25(OH)D concentrations and time since supplementation. Specifically, the exponential model was defined as follows:

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BMI, and exit BMI. Baseline concentrations of serum 25(OH)D (in ng/mL) were significantly different between groups (African American compared with white: 24.1 and 37.2, respectively; \( P = 0.01 \)). Baseline and exit concentrations of 25(OH)D were also different within each group (African American: 24.1 compared with 67.7; \( P < 0.0001 \); white: 37.2 compared with 67.3; \( P < 0.0001 \)). After only 2 mo of supplementation, differences in circulating concentrations of vitamin D between African American and white subjects became nonsignificant (mean \( \pm \) SD: 51.43 \( \pm \) 10.49 ng/mL in African Americans and whites, respectively; \( P = 0.20 \)); As shown in Figure 1, subjects with the lowest baseline circulating concentrations of vitamin D had the largest increases by 2 mo. These results are consistent with the concept that the rate of 25-hydroxylation of vitamin D3 decreases dramatically with higher initial circulating 25(OH)D concentrations. This has been previously reported in 2 recent studies in which production of 25(OH)D decreased when circulating 25(OH)D concentrations approached 40 ng/mL, probably through feedback inhibition (25, 26). The effects over time of 1 y of supplementation on serum concentrations of 25(OH)D in 12 African American and 35 white subjects are shown in Figure 2. The 2 groups were essentially indistinguishable after 1 y of vitamin D3 supplementation (67.7 compared with 67.3 ng/mL in African Americans and whites, respectively; \( P = 0.97 \)). The exponential model for African American and white subjects differed in the increment to equilibrium produced by a given constant input (parameter \( a \): 42.1 compared with 31.7 in African Americans and whites, respectively) but had similar values for the rate constant (parameter \( k \): 0.55 compared with 0.50 in African Americans and whites, respectively) representing the proportion of total mass of 25(OH)D metabolized per day. Model estimates, their estimated dispersion parameter, and 95% credible intervals are provided in Table 2. A full representation of the posterior distributions of \( a \) and \( k \) parameters by race is also provided as supplemental material (see Figure S1 under “Supplemental data” in the online issue). These results are consistent with those reported by Heaney et al (24).

No adverse events linked to vitamin D3 supplementation at 4000 IU/d were observed, and no subject reported any symptoms associated with supplementation. Six of 47 subjects had a urinary calcium:creatinine ratio of >0.2 at enrollment before starting supplementation. Five of these 6 subjects had a decrease in the ratio or remained stable after 1 y of supplementation. In one subject who had significant cardiometabolic comorbidities,
the ratio increased from 0.28 to 0.39. This subject expressed no complaints over the time of the study.

DISCUSSION

Our clinical observations of extensive hypovitaminosis D among African American males are entirely consistent with the results of the NHANES 2001–2004, which showed that >90% of African Americans have 25(OH)D serum concentrations <30 ng/mL (5). Supplementation with 4000 IU/d for 1 y elevated circulating 25(OH)D to a range (40–70 ng/mL of serum) that was recently recommended by the Endocrine Society Practice Guidelines (27). The results of this interventional study show the feasibility and efficacy of this approach in eliminating hypovitaminosis D, which is a crucial health disparity among African Americans. In addition, this regimen erased previously significant differences in serum concentrations of 25(OH)D between African American and white subjects. Furthermore, it is important to point out the absence of any significant toxicity of the vitamin D3 intervention used in this clinical study. The Endocrine Society Practice Guidelines, compared with those of the Food and Nutritional Board of the Institute of Medicine (28, 29), recommend a 2- to 3-fold increase in vitamin D3 intake, with a tolerable upper intake level of 10,000 IU/d (27). The Institute of Medicine report also concluded that circulating 25(OH)D concentrations were unrelated to various neoplasias, and in fact higher concentrations of circulating 25(OH)D could make the cancer worse. These conclusions of the Institute of Medicine report are not supported by our current data (30).

Although it is possible that optimal concentrations of circulating vitamin D are different between African Americans and whites, the results of our interventional study clearly indicate that after 1 y of vitamin D3 supplementation at 4000 IU/d the 2 groups were essentially indistinguishable on the basis of serum concentrations of 25(OH)D. These results also suggest that African American men may be able to convert vitamin D3 into 25(OH)D more rapidly than white men, as originally proposed by Heaney et al (24). However, we acknowledge that the study was not planned with the estimation of the exponential model. As a result, the inferences should be made with caution, with more emphasis provided on the point estimates of the parameters and their interpretation than on statistical significance. In addition, the credible intervals are helpful in obtaining a good understanding of a reasonable range of true values for the parameters based on the data collected in this study.

Vitamin D3-based interventions targeting adult African Americans could have a beneficial impact on the current epidemics of cardiometabolic disorders in this underserved population, would provide strong evidence for the causal role played by vitamin D deficiency in these pathologies, and could help eliminate well-documented health disparities. Some of the answers will come from prospective, randomized clinical studies.

In the past few years there has been a steady stream of news reports extolling the beneficial effects of the “sunshine vitamin” and the negative health consequences of hypovitaminosis D in the general population. These reports tend to following publication of an ever-increasing number of scientific reports showing that hypovitaminosis D has reached epidemic proportions and that lower concentrations of circulating vitamin D have an inverse correlation with the prevalence and severity of a range of diseases including diabetes and cardiovascular disease (13, 31–33). Possible explanations for the higher prevalence of diabetes and cardiovascular disease among African Americans include socioeconomic, genetic, and environmental/dietary factors (34). We propose that these health disparities are also the result of widespread hypovitaminosis D within this population and that, through appropriate vitamin D3 supplementation, vitamin D deficiency can be easily remedied in African Americans. Through well-designed prospective, randomized clinical trials it will be possible to establish whether, by using an intervention strategy that is extremely cost-effective and easy to implement, vitamin D3 supplementation has the potential to greatly reduce health disparities among African Americans.

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The authors’ responsibilities were as follows—SG-C and MSK designed the study; EG-M: analyzed the data and performed the statistical analyses; SG-C and EG-M: wrote the manuscript; and CLW and BWH: provided essential reagents and were responsible for measuring circulating vitamin D. All of the authors read and approved the final manuscript. BWH is an academic consultant to the DiaSorin Corporation. The other authors declared no conflicts of interest.

REFERENCES