

## Efficacy and safety of vitamin D<sub>3</sub> intake exceeding the lowest observed adverse effect level<sup>1-3</sup>

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### ABSTRACT

**Background:** The Food and Nutrition Board of the National Academy of Sciences states that 95  $\mu\text{g}$  vitamin D/d is the lowest observed adverse effect level (LOAEL).

**Objective:** Our objective was to assess the efficacy and safety of prolonged vitamin D<sub>3</sub> intakes of 25 and 100  $\mu\text{g}$  (1000 and 4000 IU)/d. Efficacy was based on the lowest serum 25-hydroxyvitamin D [25(OH)D] concentration achieved by subjects taking vitamin D<sub>3</sub>; potential toxicity was monitored by measuring serum calcium concentrations and by calculating urinary calcium-creatinine ratios.

**Design:** Healthy men and women ( $n = 61$ ) aged  $41 \pm 9$  y ( $\bar{x} \pm \text{SD}$ ) were randomly assigned to receive either 25 or 100  $\mu\text{g}$  vitamin D<sub>3</sub>/d for 2–5 mo, starting between January and February. Serum 25(OH)D was measured by radioimmunoassay.

**Results:** Baseline serum 25(OH)D was  $40.7 \pm 15.4$  nmol/L ( $\bar{x} \pm \text{SD}$ ). From 3 mo on, serum 25(OH)D plateaued at  $68.7 \pm 16.9$  nmol/L in the 25- $\mu\text{g}/\text{d}$  group and at  $96.4 \pm 14.6$  nmol/L in the 100- $\mu\text{g}/\text{d}$  group. Summertime serum 25(OH)D concentrations in 25 comparable subjects not taking vitamin D<sub>3</sub> were  $46.7 \pm 17.8$  nmol/L. The minimum and maximum plateau serum 25(OH)D concentrations in subjects taking 25 and 100  $\mu\text{g}$  vitamin D<sub>3</sub>/d were 40 and 100 nmol/L and 69 and 125 nmol/L, respectively. Serum calcium and urinary calcium excretion did not change significantly at either dosage during the study.

**Conclusions:** The 100- $\mu\text{g}/\text{d}$  dosage of vitamin D<sub>3</sub> effectively increased 25(OH)D to high-normal concentrations in practically all adults and serum 25(OH)D remained within the physiologic range; therefore, we consider 100  $\mu\text{g}$  vitamin D<sub>3</sub>/d to be a safe intake. *Am J Clin Nutr* 2001;73:288–94.

**KEY WORDS** Cholecalciferol, calcidiol, vitamin D<sub>3</sub>, hypercalciuria, toxicity, lowest observed adverse effect level, LOAEL, efficacy

### INTRODUCTION

Food and Nutrition Board guidelines specify 50  $\mu\text{g}/\text{d}$  as the highest vitamin D intake that healthy adults can consume without risking hypercalcemia [it is the upper limit, or the no adverse effect level (NOAEL)]. A prolonged intake of 95  $\mu\text{g}$  vitamin D/d is said to be the lowest observed adverse effect level (LOAEL), a dosage that causes hypercalcemia in healthy adults (1). These intake limits have changed little from previous guidelines (2).

However, the current guidelines (1) are based on the data of Narang et al (3), who reported that mean serum calcium concentrations were abnormally high in 6 healthy subjects who consumed 95  $\mu\text{g}$  vitamin D/d for 3 mo. More recently, Adams and Lee (4) reported a high urinary calcium-creatinine ratio in 4 patients taking nutritional supplements containing vitamin D<sub>2</sub> (ergocalciferol). Substantial concern has been expressed about the safety of consuming vitamin D at dosages greater than the highest dosage available without prescription (25  $\mu\text{g}/\text{d}$ ) (1, 5).

Directly related to this issue is the question of how much vitamin D is needed to ensure target serum 25-hydroxyvitamin D [25(OH)D] concentrations. According to the recommended dietary allowances, persons should achieve “levels of intake of essential nutrients considered. . . to be adequate to meet the known nutritional needs of practically all healthy persons” (2, 6). Serum 25(OH)D is the appropriate index of vitamin D nutritional adequacy (1). Therefore, the nutritional need for vitamin D would be the amount of it needed to ensure for “practically all healthy persons” that serum 25(OH)D concentrations are maintained above a concentration considered adequate. Moderate vitamin D malnutrition is based on what is now well documented—an inverse relation between serum 25(OH)D and parathyroid hormone (PTH) concentrations (7–9). 25(OH)D concentrations <40–50 nmol/L are considered to be insufficient (10–12). Because the suppression of PTH is seen as beneficial for bone, many now regard serum 25(OH)D concentrations  $\geq 75$ –100 nmol/L as desirable. This is the concentration at which PTH approaches a minimum in its relation with 25(OH)D (7, 9, 11, 13–15).

An intake of  $\geq 100$   $\mu\text{g}$  vitamin D<sub>3</sub> (cholecalciferol)/d may be required to ensure desirable 25(OH)D concentrations (16). However, because such intakes exceed the LOAEL, it is not feasible

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**TABLE 1**  
Characteristics of the 2 groups of subjects<sup>1</sup>

	Vitamin D <sub>3</sub> dosage	
	25 µg/d	100 µg/d
Number of subjects	33	28
Men (%)	30.3 [10]	35.7 [10]
Women (%)	69.7 [23]	64.3 [18]
Age (y)	41.6 (18–53)	39.9 (23–56)
Body weight (kg) <sup>2</sup>	67.8 ± 11.9	66.4 ± 12.9
Race		
White (%)	66.6 [22]	71.4 [20]
Black (%)	6.1 [2]	10.7 [3]
Asian (%)	27.3 [9]	17.9 [5]
Basal 25(OH)D (nmol/L) <sup>2</sup>	43.3 ± 16.8	37.9 ± 13.4
Subjects with basal 25(OH)D < 40 nmol/L (%)	36.3 [12]	57.1 [16]
Subjects with basal 25(OH)D < 25 nmol/L (%)	12.1 [4]	21.4 [6]

<sup>1</sup>Range in parentheses; *n* in brackets 25(OH)D, 25-hydroxyvitamin D.<sup>2</sup> $\bar{x} \pm SD$ .

to use them in studies of healthy adults, especially in long-term studies designed to evaluate health effects. Ethical review panels may be hesitant to approve the use of a dosage that exceeds the LOAEL, funding agencies may be hesitant to provide the funds needed for such study, and study subjects themselves may have or develop reservations that could lead to poor study compliance.

There are few data from which to establish vitamin D safety and toxicity limits. Some studies that might be considered relevant because they include data on serum calcium, urinary calcium, or both have major shortcomings, eg, ≤6 subjects (3, 4, 17), follow-up times ≤3 mo (3, 18, 19), nonspecification of the form of vitamin D used (vitamin D<sub>2</sub> or vitamin D<sub>3</sub>) (3, 4), or nonverification of the accuracy of the stated dose (3, 4). The objectives of the present study were to assess the efficacy of high vitamin D intakes, in terms of the serum 25(OH)D concentrations ensured for practically all adults, and to monitor the long-term safety of vitamin D<sub>3</sub> intakes that exceed the LOAEL.

## SUBJECTS AND METHODS

### Subjects

The study protocol was approved by an ethical review committee at the University of Toronto and subjects signed a form indicating their informed consent. We recruited 73 generally healthy volunteers, most of whom worked in the clinical laboratory departments of 2 Toronto hospitals (latitude 43°N). The characteristics of the subjects are summarized in **Table 1**. Subjects were randomly assigned to receive a vitamin D<sub>3</sub> intake of either 25 or 100 µg/d; only data for subjects who consumed vitamin D<sub>3</sub> for ≥1 mo are provided. The doses were assigned randomly by distributing the 2 dose formats in equal numbers through a partitioned rack from which each subject's first coded vial was taken, in sequence, without the giver's or the subject's knowledge of the dose it contained. Of the 73 subjects enrolled initially, 61 completed ≥1 mo of the protocol. The study began between January and February (baseline, time 0). At 0, 0.5, 1, 2, 3, 4, and 5 mo of vitamin D<sub>3</sub> supplementation, a morning urine sample (second void of the day) was collected and one tube of blood was collected to prepare serum for biochemical testing. At

the time of each blood sampling, the vials that had contained the vitamin D<sub>3</sub> solutions were collected to monitor compliance and subjects were given fresh vials. Subjects were free to withdraw from the study at any time and their withdrawal is reflected in the number of points shown in the figures. When insufficient serum samples were available from the clinical service laboratory, 25(OH)D concentrations were not measured; the measurement of calcium was given priority. Blood was collected from 25 laboratory coworkers who did not take part in the vitamin D intake study; this group served as an end-of-study reference group.

### Materials

Vitamin D<sub>3</sub> was purchased in crystalline form from Sigma (St Louis) and dissolved in US Pharmacopoeia-grade ethanol. The molar concentration of vitamin D<sub>3</sub> was adjusted to 433 µmol/L [100 µg (4000 IU) per 0.6-mL dose], which was based on an absorbance at 265 nm [7.90 absorbance units (AU) with use of an extinction coefficient of 18 300 AU · mol<sup>-1</sup> · L<sup>-1</sup>] on an 8452A diode array spectrophotometer (Hewlett-Packard, Palo Alto, CA). The lower dose was adjusted to 108 µmol/L [25 µg (1000 IU) per 0.6-mL dose]. Appropriately blanked ultraviolet absorption spectra of the doses taken before, during, and after the study remained identical. In addition, chromatographic analysis of the dose preparations consistently indicated purity by showing only the one peak appropriate for vitamin D<sub>3</sub>. Vitamin D<sub>3</sub> was consumed daily by each subject by mixing 0.6 mL of the ethanolic solution into juice or water just before drinking it (20).

### Assays

Serum 25(OH)D was measured by radioimmunoassay (DiaSorin, Stillwater, MN). Serum calcium and phosphate and urinary calcium, phosphate, and creatinine were measured with an Integra automatic chemistry analyzer (Roche, Basel, Switzerland). Urinary calcium excretion was assessed as a ratio of urinary calcium to creatinine concentrations (4, 21).

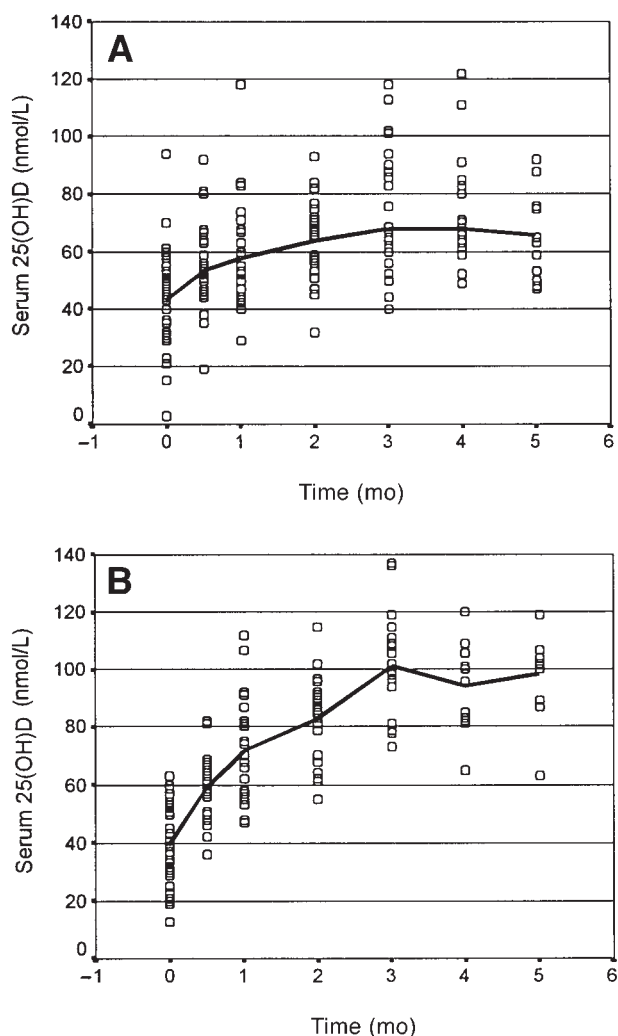
### Statistical analyses

Safety and efficacy were assessed by using repeated-measures analysis of variance (ANOVA) followed by Dunnett's multiple (pairwise) comparison *t* test to determine the CI for the mean differences from baseline at each time point (release 6.12; SAS Statistical Software, Cary, NC). A two-tailed *P* value < 0.05 was considered significant. For regression lines plotted nonparametrically, we used the locally weighted regression and smoothing scatterplot (LOWESS) approach (22) using SPSS statistical software (versions 8 or 10; SPSS Inc, Chicago). This software was also used for paired *t* test comparisons with baseline values.

### Criteria for safety and efficacy

The efficacy of a given dose of vitamin D<sub>3</sub> was based on the final serum 25(OH)D concentration (not its change). A dose was considered effective if it ensured a serum 25(OH)D concentration ≥75 nmol/L (7, 9, 11, 13–15). A dose was considered safe if all of the following criteria were met:

- 1) a mean serum calcium concentration ≤2.75 mmol/L (11 mg/dL) during vitamin D supplementation, the criterion used by others to support the current LOAEL (1);
- 2) no increase in the relative number of subjects with hypercalcemia relative to baseline, when the subjects were tested without vitamin D;



**FIGURE 1.** Serum 25-hydroxyvitamin [25(OH)D] concentrations in healthy adults before and during supplementation with 25 (A) and 100 (B)  $\mu\text{g}$  vitamin  $\text{D}_3/\text{d}$ . The heavy lines represent the locally weighted regression and smoothing scatter plot. Of 33 subjects starting the protocol at 25  $\mu\text{g}/\text{d}$ , 26 completed  $\geq 3$  mo and 15 completed the entire 5 mo of supplementation. Of 28 subjects starting the protocol at 100  $\mu\text{g}/\text{d}$ , 25 completed  $\geq 3$  mo and 15 completed the entire 5 mo of supplementation.

- 3) a mean urinary calcium-creatinine ratio  $\leq 1.0$  (when calcium and creatinine are measured in mmol;  $\leq 0.37$  when measured in mg) during vitamin D supplementation; and
- 4) no increase in the relative number of subjects with hypercalciuria relative to baseline, when the subjects were tested without vitamin D.

## RESULTS

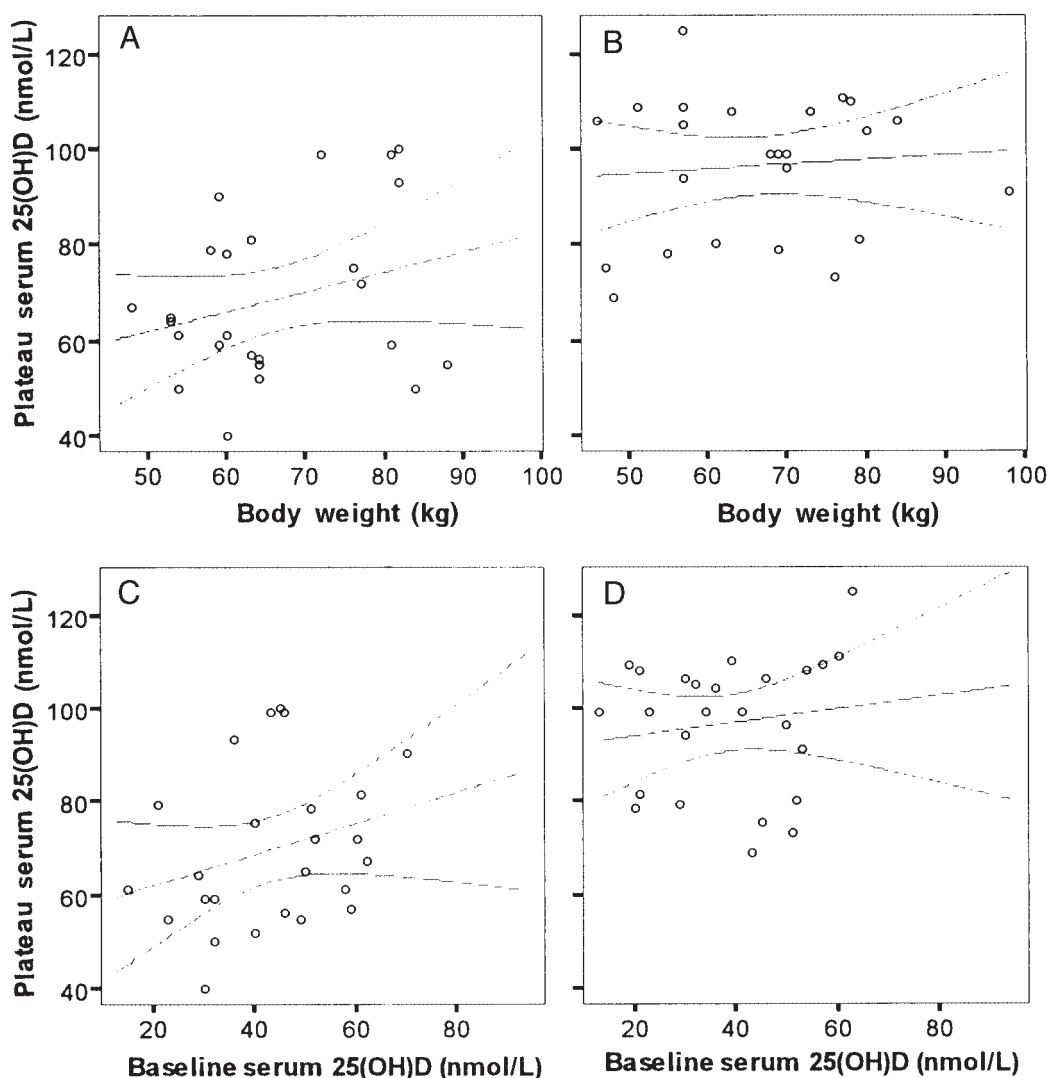
The subjects' mean ( $\pm$ SD) winter serum 25(OH)D concentration before supplementation was  $40.7 \pm 15.4$  nmol/L. The summer serum 25(OH)D concentration in 25 comparable subjects not taking vitamin  $\text{D}_3$  was  $46.7 \pm 17.8$  nmol/L. Of the 61 subjects, 28 (45.9%) had low 25(OH)D concentrations ( $<40$  nmol/L) and 10 (16.4%) had concentrations in the osteomalacic range ( $<25$  nmol/L).

Serum 25(OH)D concentrations during the supplementation protocol are shown in **Figure 1**. With the 100- $\mu\text{g}/\text{d}$  dosage, repeated-measures ANOVA and post hoc comparisons with Dunnett's test indicated that serum 25(OH)D increased significantly beginning at 2 wk until the end of the study. With the 25- $\mu\text{g}/\text{d}$  dosage, repeated-measures ANOVA and post hoc comparisons with Dunnett's test indicated a significant increase from only 1 mo on. A conventional paired  $t$  test showed the expected significant increase in 25(OH)D at 2 wk ( $P < 0.01$ ). At 3 mo, serum 25(OH)D concentrations peaked at  $68.7 \pm 16.9$  nmol/L in the 25- $\mu\text{g}/\text{d}$  group and at  $96.4 \pm 14.6$  nmol/L in the 100- $\mu\text{g}/\text{d}$  group and remained relatively stable at these concentrations for the remainder of the study. For each group, the final 25(OH)D concentration attained was not significantly affected by either the initial 25(OH)D concentration or by body weight (**Figure 2**). From 3 mo on, the minimum and maximum of the plateau serum 25(OH)D concentrations were 40 and 100 nmol/L in the 25- $\mu\text{g}/\text{d}$  group and 69 and 125 nmol/L in the 100- $\mu\text{g}/\text{d}$  groups. The 25- $\mu\text{g}/\text{d}$  dosage was effective at ensuring 25(OH)D concentrations of  $\geq 75$  nmol/L in 8 of the 23 (35%) subjects. The 100- $\mu\text{g}/\text{d}$  dosage was effective at ensuring 25(OH)D concentrations of  $\geq 75$  nmol/L in 22 of the 25 (88%) subjects.

The serum calcium concentrations and urinary calcium-creatinine ratios measured during the study are shown in **Figure 3**. In all subjects in the 25- and 100- $\mu\text{g}/\text{d}$  groups, serum calcium concentrations remained within the reference range (2.2–2.6 mmol/L). There was no significant change from baseline in serum calcium at any time by repeated-measures ANOVA. CIs for the differences between serum calcium concentrations during the study and baseline values were consistently  $\leq 0.10$  mmol/L. The upper 97.5% confidence limits for the mean difference in serum calcium results are illustrated in Figure 3 and they show with statistical confidence ( $P < 0.05$ ) that the values were consistently  $< 2.45$  mmol/L. Similarly, there was no significant change from baseline in urinary calcium-creatinine excretion ratios at any time point by repeated-measures ANOVA. The upper confidence limits for the differences between baseline urinary calcium-creatinine excretion ratios and those during supplementation were consistently  $< 0.70$  (Figure 3). There were more urinary calcium-creatinine excretion ratios  $> 1.0$  in the 100- $\mu\text{g}/\text{d}$  group (in one subject, 2 of 6 values were  $> 1.0$  during treatment) than in the 25- $\mu\text{g}/\text{d}$  group. The relative number of occurrences of hypercalciuria across the entire follow-up period was not significantly different between the 2 dosage groups on the basis of chi-square tests. Similarly, the relative number of treated subjects with hypercalciuria was not significantly different from the number of untreated subjects (baseline) with hypercalciuria. More subjects than were used here will be required to show whether the higher dose does in fact increase the occurrence of hypercalciuria. The paired  $t$  test, which was used to compare baseline values with those during treatment, is less appropriate than is ANOVA with this study design, but is more sensitive for detecting changes from baseline. Like the ANOVA, paired  $t$  tests showed no significant changes in serum calcium concentrations or urinary calcium-creatinine ratios at any time point during the 5-mo vitamin D intake period.

## DISCUSSION

Prolonged consumption of vitamin D at a dosage of 100  $\mu\text{g}/\text{d}$  resulted in plateau 25(OH)D concentrations averaging 96 nmol/L.



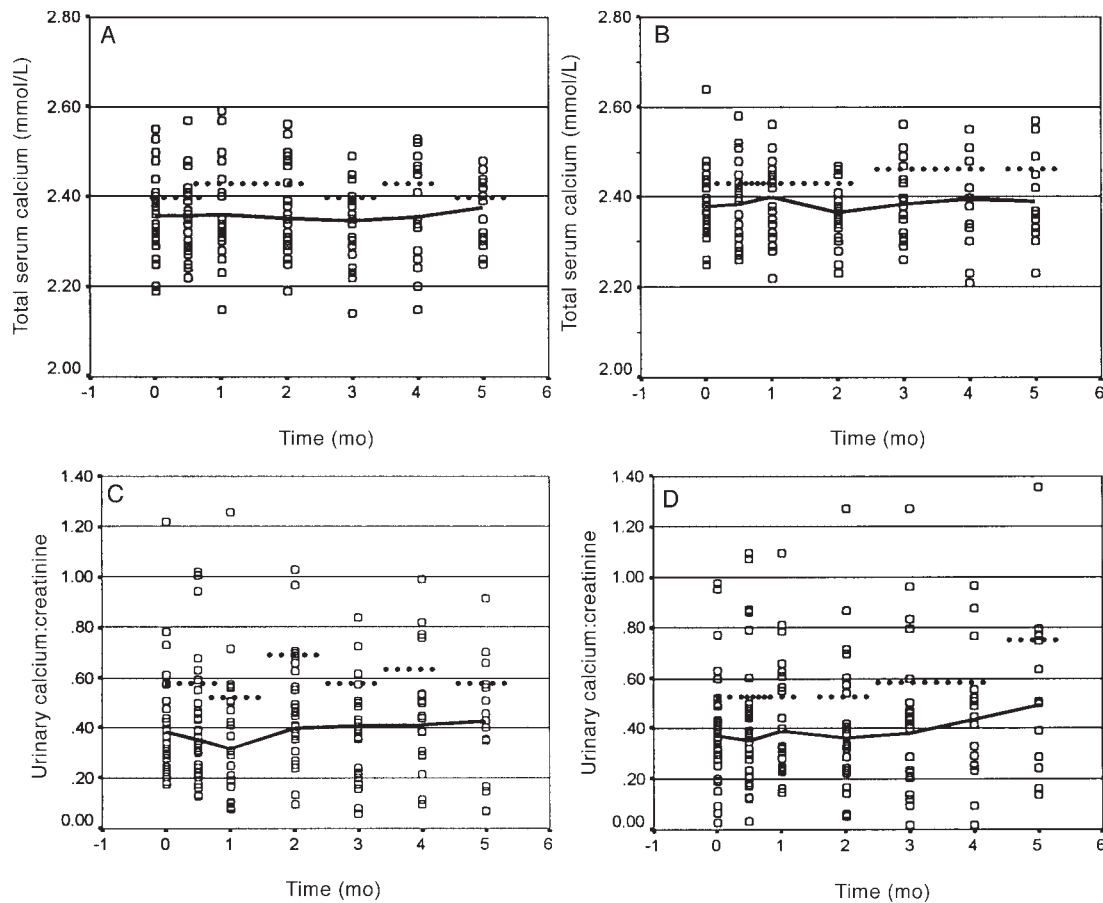
**FIGURE 2.** Effects of baseline (0 mo) body weight and serum 25-hydroxyvitamin D [25(OH)D] concentrations on final serum 25(OH)D concentrations in healthy adults after supplementation for 3–5 mo with 25 (A and C;  $n = 25$ ) or 100 (B and D;  $n = 25$ )  $\mu\text{g}$  vitamin D<sub>3</sub>/d. Because the 95% confidence limits encompass the horizontal, they indicate that plateau 25(OH)D concentrations were not significantly affected by baseline 25(OH)D concentrations or body weight (23), ie, there were no significant correlations ( $r^2$ ) for any of the correlations. A:  $y = (40.86 + 0.42) \times \text{wt}$ ;  $r^2 = 0.08$ . B:  $y = (89.89 + 0.10) \times \text{wt}$ ;  $r^2 = 0.01$ . C:  $y = (55.36 + 0.33) \times \text{baseline 25(OH)D}$ ;  $r^2 = 0.08$ . D:  $y = (90.91 + 0.15) \times \text{baseline 25(OH)D}$ ;  $r^2 = 0.02$ .

On the basis of current nutritional guidelines, one would expect 25(OH)D concentrations much higher than this because of the presumption that the 95- $\mu\text{g}/\text{d}$  LOAEL will raise the average serum calcium concentration to the hypercalcemic range ( $>2.75$  mmol/L, or 11 mg/dL) (1). In the present study, mean serum calcium consistently remained  $<2.45$  mmol/L. The vitamin D produced in the skin as a result of sun exposure is not enough to cause hypercalcemia, even though sunshine can raise circulating concentrations of 25(OH)D to  $>200$  nmol/L (16, 24). The present results expand on earlier work in which serum calcium was not affected significantly by 2-mo intakes of 100 (18) or 1250 (19)  $\mu\text{g}$  vitamin D<sub>3</sub>/d or 12-mo intakes of 350  $\mu\text{g}$  vitamin D<sub>2</sub>/d (17). Together, the evidence is overwhelming that the hypercalcemia evoked by Narang et al (3) after 3 mo of treatment with 95  $\mu\text{g}$  vitamin D/d must have been in error. We contend that without data on serum 25(OH)D concentrations, the hypercalcemia observed by Narang

et al was effectively a biological response indicating that they had grossly underestimated the amount of vitamin D in the doses they used. It is unfortunate that the study by Narang et al remains the only study cited by the Food and Nutrition Board to support the current LOAEL of 95  $\mu\text{g}/\text{d}$  (1).

The results of the present study differ slightly from those of earlier studies in that we did not detect an effect of treatment on urinary calcium excretion. Our urine samples were collected from nonfasting subjects at the second void of the morning (collected before 1000). Collection times in other studies were not specified and the subject populations may have differed from ours. Tjellesen et al (18) used the same dosage we did and showed small but significant increases in urinary calcium-creatinine excretion ratios. Arthur et al (17) reported a 50% increase in urinary calcium-creatinine ratios in 6 women who took 350  $\mu\text{g}$  vitamin D<sub>2</sub>/d for 16 mo (17). Adams and Lee (4) reported 4 cases of





**FIGURE 3.** Total serum calcium concentrations and urinary calcium-creatinine excretion ratios at baseline (0 mo) and during supplementation with 25 (A and C) and 100 (B and D)  $\mu\text{g}$  vitamin  $\text{D}_3/\text{d}$ . The heavy line in each panel is the nonparametric, locally weighted regression and smoothing scatter plot. The dotted lines reflect the upper limit of the central 95% CI for the mean change. The value of each dotted line was calculated by adding the upper limit value (97.5%) for the mean change from baseline at each time point to the mean baseline value (month 0) for each group (repeated-measures ANOVA followed by Dunnett's test).

vitamin D intoxication on the basis of only increased fasting urinary calcium-creatinine ratios.

For people with serum calcium concentrations in the reference range, the implications of high-normal urinary calcium concentrations need to be understood better before this can be regarded as an adverse response. Until there is more evidence to verify the safety of intakes of 100  $\mu\text{g}$  vitamin  $\text{D}_3/\text{d}$ , it is simple and prudent to continue to monitor calcium in randomly collected urine samples. Although randomly collected urine samples can be affected by large calcium loads in the hours before collection (25, 26), 24-h collections are notoriously affected by improperly timed collections, missed urine voids, and daily variations in calcium intake. Calcium concentrations in randomly collected urine samples correlate well with calcium concentrations in 24-h urine samples (21, 27–30). The mean calcium-creatinine ratio in randomly collected urine samples from nonfasting groups of healthy subjects is  $\approx 0.40$  (0.14 when measured in mg) (29, 31, 32). A 24-h urinary calcium excretion  $> 10$  mmol/d (400 mg/d) is considered to indicate hypercalciuria (33). Graphs of regression data for the relation between random urinary calcium-creatinine ratios versus 24-h calcium excretion (21, 28) indicate that at a 24-h calcium excretion of 10 mmol/d, the nonfasting random calcium-creatinine ratio is  $\approx 1.0$  (0.37 when measured in mg). In our study there were


more urinary calcium-creatinine ratios  $> 1.0$  in the 100- $\mu\text{g}/\text{d}$  group than in the 25- $\mu\text{g}/\text{d}$  group, but the difference was not significant.

In the absence of hypercalcemia, urinary calcium excretion is a minor contributor to renal stone disease. Stronger risk factors for calcium oxalate stones include low urine volume, hyperoxaluria, and hypocitraturia. Urinary calcium is useful for classifying normocalcemic patients who already have stone disease (34), but it cannot discriminate adults who form calcium oxalate stones from those who do not form kidney stones (33, 35). Furthermore, it is highly controversial whether isolated high-normal calcium excretion contributes adversely to either stone disease or bone health (33, 35–37). Because it is impossible to prove by statistical analysis that an agent has no harmful effect, we can only conclude that an intake of 100  $\mu\text{g}$  vitamin  $\text{D}_3/\text{d}$  is safe because there was no convincing evidence of harm.

Throughout the history of vitamin D supplementation in North America, high-dose preparations of the vitamin  $\text{D}_2$  form have generally been used. Vitamin  $\text{D}_3$  is common in lower-dose regimens, but in some parts of the world vitamin  $\text{D}_2$  is the only form licensed for use. In terms of rickets prevention, research from the 1930s was inconclusive at detecting a difference in efficacy between the 2 forms of vitamin D; therefore, pharmacopoeias continue to regard vitamin  $\text{D}_2$  as being equivalent to vitamin  $\text{D}_3$ ,

even though the latter form is more effective at raising serum 25(OH)D concentrations (20). What seems to have been forgotten is that the literature of half a century ago established that, at high doses, there was a greater risk of toxicity with what was called "the purely artificial" compound, vitamin D<sub>2</sub> (38–40). One explanation for the difference in toxicity was the poorer stability and greater impurity of vitamin D<sub>2</sub> than of vitamin D<sub>3</sub> preparations (38). There are newer reasons why vitamin D<sub>2</sub> has a greater potential for harm. First, vitamin D binding protein has a weaker affinity for the vitamin D<sub>2</sub> metabolites than for 25-hydroxyvitamin D<sub>3</sub> and 1,25-dihydroxyvitamin D<sub>3</sub> (41–43). This means that the proportions of free 25-hydroxyvitamin D<sub>2</sub> and 1,25-dihydroxyvitamin D<sub>2</sub> are higher and more biologically available. Second, unique biologically active metabolites are produced from vitamin D<sub>2</sub> in humans and there are no analogous metabolites derived from vitamin D<sub>3</sub> (44). There is no doubt that vitamin D<sub>2</sub> is a synthetic analogue of vitamin D, with different characteristics. It is an anachronism to regard vitamin D<sub>2</sub> as a vitamin. Future research into the toxicology of this vitamin needs to focus on vitamin D<sub>3</sub> as being something distinct from vitamin D<sub>2</sub>, for which almost all our current toxicity data relate to.

The working definition of the recommended dietary allowance has been to ensure "levels of intake of essential nutrients considered, in the judgment of the Food and Nutrition Board on the basis of available scientific knowledge, to be adequate to meet the known nutritional needs of practically all healthy persons" (2, 6). For vitamin D, the relevant question could be, what 25(OH)D concentration is desirable and how much vitamin D is needed to ensure that most adults attain this intake (45)? We did not measure PTH in this study, so we cannot address the question of what the desirable vitamin D intake is. However, the present results do provide insights into the lowest 25(OH)D concentrations that can be reasonably ensured in adults consuming 25 and 100 µg vitamin D<sub>3</sub>/d. The 25-µg/d intake offered reasonable assurance that serum 25(OH)D concentrations in adults would be >40 nmol/L, but did not ensure that most subjects would attain serum 25(OH)D concentrations considered desirable (>75 nmol/L). The 100-µg/d intake offered reasonable assurance that 25(OH)D concentrations in adults would be >69 nmol/L (Figures 1 and 2), close to the lower end of the desirable concentration.

If the serum 25(OH)D concentration is the appropriate measure of vitamin D nutritional adequacy (1), then more of the present type of specific data are needed to define the amounts of vitamin D required to ensure that for "practically all healthy persons" serum 25(OH)D concentrations are maintained above an amount considered adequate. There are subgroups who require ≥25 µg vitamin D/d to maintain acceptable 25(OH)D concentrations. Gloth et al (46) reported that in older patients with 25(OH)D concentrations <25 nmol/L, vitamin D intakes ranged as high as 29 µg/d. Patients with cystic fibrosis require >20 µg vitamin D/d to maintain 25(OH)D concentrations >40 nmol/L (47). Despite the greater number of subjects and the longer follow-up in the present study than in previous comparable studies (3, 18, 19), consumption of vitamin D<sub>3</sub> at intakes ≥100 µg/d causes no harm and effectively raises 25(OH)D to high-normal concentrations in practically all adults. 

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