

Serum 25-hydroxyvitamin D concentrations and related dietary factors in peri- and postmenopausal Japanese women^{1,2}

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ABSTRACT

Background: Few studies of vitamin D nutrition in Asian populations have been conducted.

Objective: The objective was to assess 25-hydroxyvitamin D [25(OH)D] concentrations in healthy elderly Japanese women during the winter and to determine whether 25(OH)D concentrations are associated with lifestyle.

Design: We investigated 151 women aged 66.5 ± 6.7 y ($\bar{x} \pm$ SD) living in a rural community in February 1999. Serum 25(OH)D and intact parathyroid hormone were measured by using HPLC and an immunoradiometric assay, respectively. Information on lifestyle factors, including sunshine exposure and the consumption of vitamin D-rich foods, was also obtained through an interview.

Results: The mean (\pm SD) 25(OH)D concentration was 59.9 ± 17.0 nmol/L. Vitamin D insufficiencies (<30 nmol/L) were found in 4.6% of the women, a value lower than that found in white populations. No correlation was found between age and 25(OH)D concentrations ($r = 0.004$, $P = 0.957$). The 25(OH)D concentration of subjects who consumed fish frequently (≥ 4 times/wk) was 10.1 nmol/L higher ($P < 0.001$) than that of subjects with a moderate consumption of fish (1–3 times/wk). Additionally, those who did not consume eggs had significantly lower 25(OH)D concentrations than did those who consumed eggs ≥ 1 time/wk ($P < 0.05$).

Conclusions: The nutritional status of vitamin D in Japanese populations seems to be better than that in most Western populations. Frequent fish consumption is believed to help maintain adequate concentrations of serum 25(OH)D in elderly Japanese women during the winter. *Am J Clin Nutr* 2000;71:1161–5.

KEY WORDS Epidemiology, fish consumption, 25-hydroxyvitamin D, 25(OH)D, Japan, parathyroid hormone, winter, elderly women

INTRODUCTION

Low exposure to sunlight during the winter can sometimes lead to vitamin D insufficiencies. Low concentrations of vitamin D are particularly problematic for women, who have a greater risk of osteoporosis than do men (1–3). Both homebound and ambulatory individuals are more prone to vitamins D insufficiencies during the winter. In one report, nearly half of the ambulatory women included in the study had insufficient concentrations of serum 25-hydroxyvitamin D [25(OH)D], a form of

vitamin D that is stored in the body, during the winter (4). Insufficient 25(OH)D concentrations are known to be a risk factor for the acceleration of age-related bone loss. Although the mechanism of this accelerated bone loss is not fully understood, an elevated concentration of parathyroid hormone as a result of an insufficient 25(OH)D concentration is believed to play a role (5).

Epidemiologic information on the nutritional status of vitamin D in white populations has been accumulating, but only a few studies have been conducted in other ethnic groups. Previously, we reported on summertime 25(OH)D concentrations in a female Japanese population (6). The study showed that the average 25(OH)D concentration in the population was higher than most of the values reported for Western populations. The higher 25(OH)D concentration in the Japanese population was probably related to lifestyle factors, including the amount of time spent in the sunlight. These lifestyle factors appear to be sufficient to offset the potential disadvantage of having darker skin (7). However, no information was available on 25(OH)D concentrations in the winter or the prevalence of vitamin D insufficiencies. Therefore, the present epidemiologic study was designed to determine wintertime 25(OH)D concentrations in a Japanese population.

Although the average calcium intake is known to be lower in Japan than in the United States and Europe, the incidence of osteoporosis is also much lower in Japan than in the United States or Europe (8). Differences in lifestyle or genetic factors are believed to explain this observation (9). Conclusive evidence, however, has not been identified. An evaluation of vitamin D concentrations in the Japanese population may therefore shed some light on this matter. Thus, the objectives of the present study were to assess wintertime 25(OH)D concentrations and identify vitamin D insufficiencies in a population of healthy elderly Japanese women and to determine whether 25(OH)D concentrations are associated with lifestyle factors, including sunlight exposure and dietary vitamin D intake.

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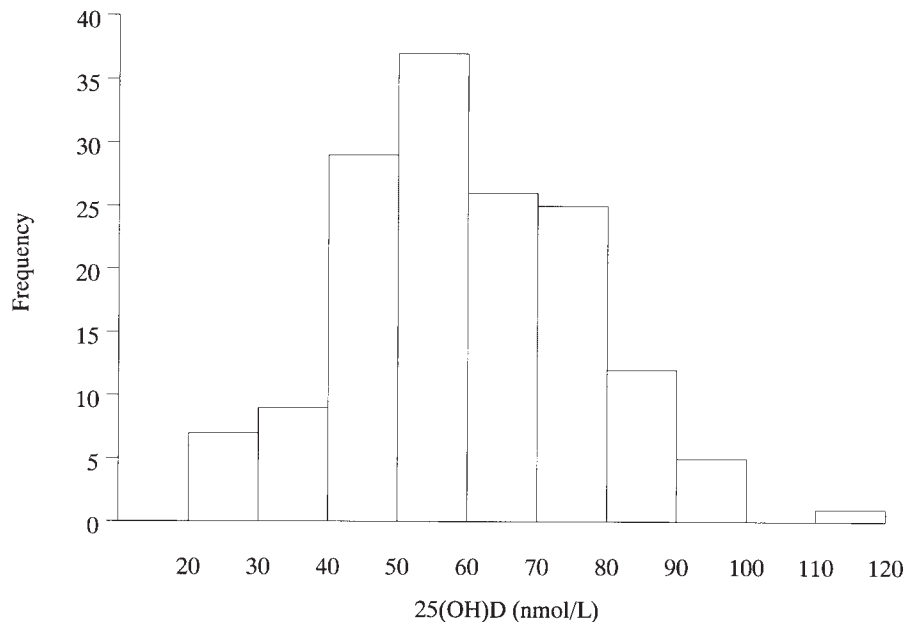


FIGURE 1. Frequency distribution of 25-hydroxyvitamin D [25(OH)D] concentrations in the winter in elderly Japanese women. The mean (\pm SD) value was 59.9 ± 17.0 nmol/L.

SUBJECTS AND METHODS

Two hundred thirty-six healthy, ambulatory female residents of a rural community in Toyosaka City, Japan (lat $37^{\circ} 54'N$), aged ≥ 40 y and who participated in a health checkup program in 1997 were contacted by mail and asked to participate in the present study; 157 took part in the study in February 1999. On the basis of the annual health checkup data from September 1998, the serum creatinine concentrations of the participants were between 26.5 and 79.6 $\mu\text{mol/L}$ (0.3 and 0.9 mg/dL), which is considered normal. There was no evidence of decreased renal function in any of the participants. Informed consent was obtained from all of the participants. The study protocol was designed according to the guidelines established by the Ethical Committee of Niigata University School of Medicine.

The exclusion criteria for this study consisted of any condition or disease that might affect vitamin D metabolism. Accordingly, 1 woman with viral hepatitis and 5 women taking vitamin D or multivitamin supplements were excluded from the study. In total, the data for 151 women were analyzed.

Demographic information—including age, previous history of disease, and occupation—was obtained from the records of the annual health checkup program. Toyosaka City is surrounded by rice fields, and many of the participants were involved in some aspect of farming. Forty-seven percent of the subjects listed farming as their occupation, but these individuals did not engage in farming activities during the winter. Information on present illnesses, medication use, vitamin and calcium supplement use, and other lifestyle factors were obtained through an interview. To evaluate individual levels of sunlight exposure, the 9-point sunshine scale [validated in previous reports (1, 10)] was used. Vitamin D–fortified foods are unavailable in Japan, so the major dietary sources of vitamin D are fish, eggs, and mushrooms (11). Information on the consumption of these foods over the previous week was obtained and categorized as none, sometimes (1–3 times/wk), and frequently

(≥ 4 times/wk). The climate in the Toyosaka City area during the winter is rainy or snowy; the average period of sunshine in January–February 1999 was only 2.3 h/d.

Blood specimens were drawn in the afternoon and stored at -80°C until analyzed. The subjects were not told to fast before the sampling. Serum concentrations of 25-hydroxyergocalciferol [25(OH) D_2 ; derived from mushrooms] and 25-hydroxycholecalciferol [25(OH) D_3 ; derived from animal products and cutaneous biosynthesis] were measured by using 2-step HPLC (12). Standard compounds of 25(OH) D_2 and 25(OH) D_3 were purchased from Tetriconics (Madison, WI). The interassay CVs were 3.4% for 25(OH) D_2 and 4.1% for 25(OH) D_3 . Intact parathyroid hormone concentrations were measured with a 2-site immunoradiometric assay (Nichols Institute Diagnostics, San Juan Capistrano, CA) (13). The interassay CV was 5.6%. The reference range for intact parathyroid hormone was 1.1–5.3 pmol/L (14).

Mean (\pm SD) values were used to represent continuous variables, such as 25(OH)D and intact parathyroid hormone concentrations and age. Comparisons between the means of 2 continuous variables were evaluated by using Student's *t* test. Pearson's product-moment correlation coefficient (*r*) was calculated to describe linear correlations between 2 continuous variables. One-way analysis of variance was used to compare mean values of > 2 groups with Bonferroni's multiple comparisons for any 2 groups. A *P* value < 0.05 was considered statistically significant. SAS (version 6.12; SAS Institute Inc, Cary, NC) was used for the analyses.

RESULTS

The mean age of the subjects was 66.5 ± 6.7 y (range: 46–82 y). The mean concentrations of 25(OH) D_2 and 25(OH) D_3 were 0.6 ± 2.7 and 59.4 ± 16.9 nmol/L, respectively. Because the concentrations of 25(OH) D_2 were much lower than those of 25(OH) D_3 , 25(OH)D will be used to denote the summation of 25(OH) D_2 and 25(OH) D_3 . The distribution of 25(OH)D

TABLE 1

Serum concentrations of 25-hydroxyvitamin D in elderly Japanese women by frequency of fish consumption¹

Frequency of fish consumption	Value
	nmol/L
Frequently, ≥ 4 times/wk ($n = 80$)	65.0 ± 14.7^2
Sometimes, 1–3 times/wk ($n = 65$)	54.9 ± 18.3
None ($n = 3$)	49.2 ± 8.5

¹ $\bar{x} \pm$ SD. Data for 3 women were not available. Values represent the sum of 25-hydroxycholecalciferol and 25-hydroxyergocalciferol.

²Significantly different from sometimes, $P < 0.001$.

concentrations is shown in **Figure 1**. The average value was 59.9 ± 17.0 nmol/L. Seven women (4.6%) had vitamin D insufficiency, defined as a 25(OH)D concentration < 30 nmol/L; 12 women (7.9%) had vitamin D insufficiency on the basis of a less conservative definition of < 37.5 nmol/L. When the 25(OH)D concentrations were plotted against age, no correlation ($r = 0.004$, $P = 0.957$) between these factors was found.

Analysis of variance showed that fish consumption was significantly associated with 25(OH)D concentrations ($P < 0.001$). A comparison of mean serum 25(OH)D concentrations by frequency of fish consumption is shown in **Table 1**. Those who consumed fish frequently had a significantly greater 25(OH)D concentration (by 10.1 nmol/L) than did those who consumed fish sometimes. A comparison of mean 25(OH)D concentrations by frequency of egg consumption is shown in **Table 2**. The group with no egg consumption had a significantly lower mean 25(OH)D concentration than did the other 2 groups, by ≈ 17 nmol/L. Consumption of mushrooms and the amount of sunshine exposure were not significantly correlated with 25(OH)D concentrations.

The average concentration of intact parathyroid hormone was 3.1 ± 1.4 pmol/L. Intact parathyroid hormone concentrations were constant (2.9 ± 1.1 pmol/L) over all ages until 75 y. After 75 y, the mean intact parathyroid hormone concentration was 4.2 ± 2.4 pmol/L, which is significantly greater than that of the age group < 75 y ($P = 0.045$). However, most subjects (148 of 151) had normal parathyroid hormone concentrations. Elevated intact parathyroid hormone concentrations (11.4 pmol/L) were found in only one subject, who had a 25(OH)D concentration of 20.5 nmol/L. In our preceding study of summertime 25(OH)D concentrations (6), however, this same subject had normal intact parathyroid hormone and 25(OH)D concentrations. Thus, this subject was judged to have hyperparathyroidism due to 25(OH)D insufficiency. Two subjects also had slightly elevated intact parathyroid hormone concentrations: 64 and 66 pmol/L.

DISCUSSION

Several epidemiologic studies reported wintertime 25(OH)D concentrations in healthy elderly females, which ranged from 19 to 61 nmol/L (2, 4, 15–21). The variability in these mean 25(OH)D concentrations was not always dependent on the latitudinal location. Rather, the reports suggest that lifestyle factors, such as attitude toward sunlight exposure, were very influential (2). Compared with previous reports, the mean 25(OH)D concentration in the present study, 59.9 nmol/L, was one of the highest concentrations ever reported. This finding was somewhat unexpected for the following reasons: there was no evidence to

support better biosynthesis of vitamin D in the Japanese population in the present study than in other white populations, the average period of sunshine was short (2.3 h/d), sunbathing was not enjoyed by this population, and darker skin appears to be less advantageous for the biosynthesis of vitamin D. Additionally, vitamin D–fortified food is not produced in Japan. Therefore, the reason for the high mean concentration of 25(OH)D in the Japanese population in the present study is uncertain.

The prevalences of vitamin D insufficiency in this population were 4.6% and 7.9% on the basis of the 2 definitions of vitamin D insufficiency (30 and 37.5 nmol/L, respectively). Previous studies of healthy elderly white women reported prevalences of vitamin D insufficiency of 14% (16), 39% (4), 47% (2), and 65% (18) on the basis of a threshold definition of 30 nmol/L, and of 15% (18) and 34% (17) on the basis of a threshold definition of 37.5 nmol/L. Vitamin D insufficiency in the present population was lower than that found in previous studies; however, the selection procedure for study participants in the present study may have been biased because participants from the health checkup program were considered to be healthier than subjects from the overall community.

Because subjects who ate fish frequently (≥ 4 times/wk) had significantly higher 25(OH)D concentrations (by an average of 10 nmol/L) than did subjects who ate fish 1–3 times/wk, the frequency of fish consumption may be an important determinant of 25(OH)D concentrations in the winter. However, this association was not found in our previous study of summertime 25(OH)D concentrations (6). The Japanese eat many kinds of fish, including raw fish, which contains 10–12 μ g (400–4800 IU) 25(OH)D₃ per 100 g fish (22), and fish seems to be a major source of their dietary vitamin D. Consequently, dietary fish intake may provide a clue in explaining the adequate concentrations of 25(OH)D in this population. Fish consumption in Japan (69.9 kg/y per capita) is much higher than it is in Europe (23.1 kg/y per capita) and the United States (21.9 kg/y per capita), and Japan has one of the highest rates of fish consumption in the world (23). This trend may be even more apparent in elderly Japanese, who tend to prefer fish over meat. In addition to fish consumption, egg consumption seemed to be another significant source of dietary vitamin D in the Japanese population in the present study. Our data suggest that a lack of egg consumption is a lost opportunity for increasing 25(OH)D concentrations.

The average age of the present population (66.5 y) was slightly less than that of populations in previous epidemiologic studies (2, 4, 15–21). However, because no correlation was found between age and 25(OH)D concentrations, the lower average age of the present population probably did not affect our results. Serum

TABLE 2

Serum concentrations of 25-hydroxyvitamin D in elderly Japanese women by frequency of egg consumption¹

Frequency of egg consumption	Value
	nmol/L
Frequently, ≥ 4 times/wk ($n = 58$)	60.9 ± 18.5^2
Sometimes, 1–3 times/wk ($n = 82$)	61.3 ± 15.7^2
None ($n = 10$)	44.2 ± 12.3

¹ $\bar{x} \pm$ SD. Data for one woman were not available. Values represent the sum of 25-hydroxycholecalciferol and 25-hydroxyergocalciferol.

²Significantly different from none, $P < 0.05$.


25(OH)D concentrations are commonly believed to decrease with age. However, the present study and 2 previous studies (6, 24) provided no evidence to confirm this association. In fact, these studies suggested that there is no age-related decline in serum 25(OH)D concentrations in healthy populations, even in the winter.

Although 2 conservative definitions of vitamin D insufficiency were used in the present study [25(OH)D concentrations <30 and <37.5 nmol/L], the 25(OH)D concentration at which serum parathyroid hormone concentrations begin to rise is controversial. Researchers have suggested that the threshold may be as high as 50 (25), 78 (16), or 110 nmol/L (26). In the present study, only one subject—whose 25(OH)D concentration was 20.5 nmol/L—was considered to have an elevated parathyroid hormone concentration as a result of vitamin D insufficiency. Parathyroid hormone concentrations were satisfactory in all other subjects, even in those with 25(OH)D concentrations between 25 and 37.5 nmol/L. This finding suggests that the threshold 25(OH)D concentration for vitamin D insufficiency may be lower than the usually quoted value of 37.5 nmol/L. Meanwhile, 2 subjects with 25(OH)D concentrations of 57.6 and 69.0 nmol/L appeared to have slightly elevated parathyroid hormone concentrations (ie, intact parathyroid hormone concentrations of 64 and 66 pmol/L, respectively) that exceeded the reference value. This finding may have been related to the age of the subjects (77 and 80 y, respectively). However, the results of the present study do not clarify whether the elevated parathyroid hormone concentrations were related to the 25(OH)D concentration or to the advanced age of the subjects.

The adequate concentration of serum 25(OH)D in the present population seems to be in line with the lower incidence of osteoporosis in the Japanese population. The incidence of osteoporosis varies internationally (27) and is 33–50% lower than the incidence in the United States and northern Europe, even though the intake of calcium is lower in Japan than it is in these other 2 areas (9). Fujita (9) discussed this issue in relation to various lifestyle aspects of the Japanese, but did not mention the nutritional status of vitamin D. Insufficient 25(OH)D concentrations resulting in hyperparathyroidism increase the risk of bone loss by increasing the rate of bone turnover. Thus, we believe that the high serum 25(OH)D concentrations in this population may be an important reason for the low incidence of osteoporosis in Japan.

The present epidemiologic study, which targeted a population of healthy elderly Japanese women during the winter, yielded the following findings:

- 1) The mean 25(OH)D concentration of the population was more adequate than concentrations reported for most white populations.
- 2) The proportion of women with a 25(OH)D insufficiency was less than that found in studies of white populations.
- 3) Fish consumption was positively associated with serum 25(OH)D concentrations in elderly Japanese women.
- 4) Consumption of no eggs may increase the risk of low 25(OH)D concentrations.

These findings suggest that increased fish consumption is helpful in maintaining adequate concentrations of serum 25(OH)D during the winter. Future studies should use analytic techniques to clarify the effects of dietary vitamin D on serum 25(OH)D concentrations. 

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