Original Article

A Cross Sectional Study of Serum Vitamin D Levels in Perimenopausal and Postmenopausal Women Working Outdoors or Indoors

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Objective: To explore the serum vitamin D levels in perimenopausal and postmenopausal women living in Chiang Mai, Thailand, and compare the level of vitamin D between women working outdoors and indoors.

Materials and Methods: A cross-sectional study was performed. All participants resided in Chiang Mai, Thailand, and were divided into two groups by occupation characteristics. One group was women working outdoors, the other group were women working indoors. Blood collection was done in every subject for levels of vitamin D as 25-hydroxy vitamin D or 25(OH)D, BUN, creatinine, AST, ALT, calcium, and albumin.

Results: Eighty women were recruited. Two women in the indoor group were excluded due to abnormal liver enzymes (one woman) and abnormal renal function tests (one woman). Therefore, 38 women were in the indoor group and 40 women in the outdoor group. The mean serum 25(OH)D for all participants was 27.9±8.7 ng/mL, in which 29.3±8.6 ng/mL for the outdoor group and 26.4±8.6 ng/mL for the indoor group (p=0.130). The prevalence of vitamin D insufficiency based on 25(OH)D <30 ng/mL was 57.5% and 77.5% for the outdoor group and the indoor group, respectively. While the prevalence of vitamin D deficiency based on 25(OH)D <20 ng/mL was 12.5% and 25% for the outdoor group and indoor group, respectively. In which the prevalence of vitamin D insufficiency and deficiency in both groups had no statistical significance. The serum 25(OH)D of the participants who used sunscreen were lower than those who did not, with no statistical significance as 26.3±6.4 ng/mL and 29.2±10.6 ng/mL.

Conclusion: Serum vitamin D levels in perimenopausal and postmenopausal women in Chiang Mai were low and the prevalence of vitamin D insufficiency was high in both groups. There was no significant difference between women who work outdoors and indoors. Therefore, menopausal women who work outdoors are still a high-risk group for vitamin D insufficiency.

Keywords: Vitamin D; 25(OH)D; Outdoors; Indoors; Perimenopausal women; Postmenopausal women

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Vitamin D is one of the most important vitamins for the human body as it increases calcium absorption to maintain bone strength and prevent fractures(1). Moreover, it is a crucial factor to improve muscular strength. Reduction of the vitamin D levels leads to various conditions, including diabetes, hypertension, immune disease, infection, cardiovascular disease, and cancer(2-5).

The skin level of the stratum granulosum of the human body contains provitamin D3 or 7-dehydrocholesterol. When skin is exposed to sunlight that contains Ultraviolet B radiation (UVB), it promotes the production of vitamin D. When UVB at a wavelength of 290 to 320 nm exposes and penetrates skin layers within two hours, then provitamin D3 transforms to previtamin D3. After that, previtamin D3 is isomerized and transformed to vitamin D3 at the skin and then binds with protein to enter the blood circulation and synthesize at the hepatic and renal sites to create an active form of vitamin D or 25-hydroxy vitamin D [25(OH)D] or calcidiol(1,4). However, sources of vitamin D can be found not only in natural body synthesis but also in diet and supplements that are in the form of vitamin D2 (ergocalciferol). Vitamin D deficiency can reduce serum calcium and result in osteoporosis(6).

Studies measured vitamin D levels by evaluating serum 25(OH)D or calcidiol. The prevalence of vitamin D insufficiency based on 25(OH)D <30 ng/mL in the South, Central, North, and Northeast regions of Thailand were 43.8%, 43.1%, 39.1%, and 34.2%, respectively(7). While the prevalence of vitamin

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D deficiency based on 25(OH)D <20 ng/mL in the elderly in Bangkok, the Capital of Thailand, was 31.8%[8]. Previous studies found a higher prevalence of vitamin D deficiency in the older participants rather than the younger, due to less sun exposure or decrease vitamin D synthesis at the skin[9,10]. Another study in Hawaii revealed that the average serum 25(OH)D level in people with regular sun exposure was 31.6 ng/mL and the prevalence of vitamin D insufficiency was 51%. While participants with sufficient vitamin D, the highest serum vitamin D level detected was 62 ng/mL[11]. The study of vitamin D status and prediction of hypovitaminosis D in Italian children and adolescents revealed that the prevalence of vitamin D insufficiency and deficiency was 33.6% and 45.9%, respectively. The prevalence of participants with sufficient vitamin D was only 20.5%[12]. From the published papers above, the authors learned that a high percentage of vitamin D insufficiency occurs in countries whether they are near or far from the equator. The risk factors for vitamin D insufficiency and deficiency were increased age at above 50 years, female, dark skin pigmentation, less sun exposure, sunscreen usage, winter season, obesity with a body mass index greater than 30, malabsorption, renal dysfunction, and decrease intake of vitamin D[12-14].

Menopause is a consequence of the decrease of ovarian function, leading to less production of hormones, especially estrogen. Estrogen deficiency leads to diseases in the cardiovascular, genital, and skin systems. The lack of the aforementioned hormones accelerates the loss of bone mass rather than remodeling[15]. Changes in microarchitecture can affect bone strength as it tends to fracture easily. Around the age of 80, the trabecular bone will have deteriorated by 50%. The tendency of the postmenopausal and aging population will also be raised, and the higher rate of osteoporosis and fracture will also be increased.

A study in Khon Kaen, a province in Northeast Thailand, showed the prevalence of Vitamin D deficiency among postmenopausal women with a mean serum level of 25(OH)D at 32.58 ng/mL. The level to define deficiency was 35 ng/mL or less because this level is significantly associated with a rising of parathyroid hormones. Furthermore, the older women who live in rural areas had a lower rate of vitamin D deficiency than in the urban areas at 15.4% and 65.4%, respectively[16].

Postmenopausal women are a high-risk group for vitamin D deficiency and osteoporosis. So, it is very important to explore in terms of epidemiology, diagnosis, prevention, and treatment for vitamin D deficiency. Sunlight and vitamin D have close relationships with each other. Inadequate sun exposure relates to vitamin D deficiency and is reflected by the low level of serum vitamin D. Limited studies of serum vitamin D levels exist in menopausal women with or without sun exposure. Therefore, the present study was designed to compare the serum vitamin D levels in perimenopausal or postmenopausal women between working outdoors and indoors. The results of the present study will provide information about vitamin D deficiency. If sun exposure is not helpful for an adequate serum level of vitamin D, then vitamin D supplements should be encouraged in all menopausal women independent from occupational characteristics.

Material and Methods

Study design

The authors performed a cross-sectional study by recruiting 80 perimenopausal or postmenopausal women. They were separated into two groups by working status as the outdoor and the indoor group. Participants were recruited by randomized selection from a Menopause Clinic at Maharaj Nakorn Chiang Mai Hospital, as well as Fang and Chiang Dao Hospital, two community hospitals in Chiang Mai, Thailand.

Inclusion criteria: All participants were in menopausal period based on an age of at least 40 years old. Postmenopause was defined by amenorrhea of 12 months or more in women older than 45 years, perimenopause was defined by repeated irregular cycles with vasomotor symptoms. Women younger than 45 years suspected of menopause had the serum FSH tested two times, four weeks apart, and the serum FSH level was at least 25 mIU/mL. All were in good health with no medical condition such as thyroid, parathyroid, liver, renal disease, and were not receiving any hormone that led to amenorrhea.

Exclusion criteria: Those who had Thyroid, parathyroid, liver, renal disease, and received vitamin D supplement were exclude.

None of the participants in the present study ever received vitamin D supplements, had medical conditions as mentioned above, and had diagnosis of osteoporosis. All were Thai and able to communicate in Thai. Two women in the indoor group were excluded, one due to abnormal liver enzymes as aspartate aminotransferase (AST) >40 u/L or alanine aminotransferase (ALT) >56 u/L and another one due to abnormal renal function tests with creatinine

>1.2 mg%.

Sample size calculation was based on a previous study[12], with the mean serum 25(OH)D in the sufficient group and the insufficiency group were 51.8 ng/mL and 28.2 ng/mL, respectively, and estimated 10% error. The sample size for each group was 38.

Five mL of non-fasting blood was obtained from every subject to measure the levels of 25(OH)D, blood urea nitrogen (BUN), creatinine, AST, ALT, calcium, and albumin. Albumin was required for calculating the corrected calcium.

Serum 25(OH)D levels were measured by electrochemiluminescent competitive immunoassay, using a DiaSorin LIAISON analyzer with a minimal limit detection of 2 ng/mL and the upper limit of detection of 150 ng/mL .

The present study protocol was approved by the Ethics Committee of the Faculty of Medicine, Chiang Mai University (No. 3970).

Definition of the study variables

Demographic characteristics and socioeconomic data, occupation, and sun protection were assessed by a questionnaire. Exposure to the sunlight and work hours or hours per day were assessed by a questionnaire as well. The perimenopausal stage was defined for women at the age of 40 years old or more with irregular periods such as decrease or increase of days or amount of bleeding or with spotting between periods or lack of period more than two months but not more than 12 months. The postmenopausal stage was defined as women at the age of 40 or more who lack periods of more than 12 months[17]. Diagnosis of menopause in women aged 40 to 44 years old combined menstrual history and FSH level. The indoor group was those who worked at least six hours per day in the shade, and the outdoor group was those who worked outdoors at least three hours per day between 10 a.m. to 3 p.m.

Statistical analysis

The descriptive data were expressed as mean and median. The normal distribution of variables was assessed using the Kolmogorov-Smirnov test. The differences between the two groups were assessed by Student's t-test for the mean, Mann-Whitney U test for the median, and chi-square test for proportion. The level of significance applied in the statistic tests was 5% with 95% confidence interval (CI). The IBM SPSS Statistics software, version 22.0 (IBM Corp., Armonk, NY, USA) was used for the statistical analysis.

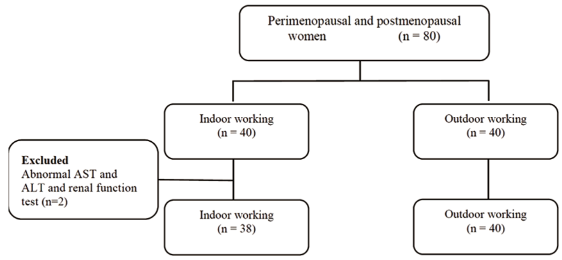

Figure 1. Diagram of number of population.

Results

Eighty perimenopausal and postmenopausal women were selected from the Menopause Clinics at Maharaj Nakorn Chiang Mai Hospital and two hospitals in the Chiang Mai, Thailand Province. The mean serum of 25(OH)D for all was 27.9±8.7 ng/mL.

When separating into two groups as in Figure 1, two in the indoor group were excluded due to abnormal liver enzyme and renal function tests. Therefore, there were 38 women in the indoor group and 40 women in the outdoor group.

Baseline characteristics of participants and baseline chemistry data in both groups are shown in Table 1 and 2. The menopausal stage of both groups was similar (p=0.306). There were more postmenopausal women than perimenopausal women in both groups at 70% for the outdoor group and 76.3% for the indoor group. The most common occupation for the outdoor group was agriculture at 72.5%, whereas the indoor group was government officers at 44.7%. The mean age of the outdoor group and indoor group was 54.3 years-old and 51.0 years-old, respectively, with statistical significance (p=0.003). The level of education between the two groups was different. Most of the outdoor women graduated from primary school at 80% while most of the indoor women graduated from universities with a bachelor's degree or higher at 55.2%. Moreover, the outdoor group had a lower monthly income than the indoor group at 93 U.S. Dollars and 781 U.S. Dollars, respectively. The work hours per day were significantly shorter in the outdoor group. Although the outdoor group was exposed to more sunlight than the indoor group, the rate of sunscreen usage was higher in the indoor group.

The mean serum 25(OH)D level in both groups was low at 29.3±8.6 ng/mL and 26.4±8.6 ng/mL in the outdoor and the indoor group, respectively (p=0.130). When vitamin D insufficiency was defined as 25(OH)D level <30 ng/mL, the prevalence of vitamin D insufficiency was high in both groups at 57.5% in

Table 1. Demographic data of participants (outdoor vs. indoors group) (n=78)

Characteristics	Outdoor (n=40)	Indoor (n=38)	p-value*
Age (year); mean±SD	54.3±4.9	51.0±4.6	0.003**
Perimenopause	49.9±4.0	49.1±2.3	
Postmenopause	56.2±4.1	51.3±5.1	
Menopausal stage (year); n (%)			0.306
Perimenopause	12 (30.0)	9 (23.7)	
Postmenopause	28 (70.0)	29 (76.3)	
Parity; median (range)	2 (0 to 4)	2 (0 to 4)	
BMI (kg/m^2); mean±SD	23.9±3.2	22.9±3.2	
Work hour per day (hour); mean±SD	6.7±1.3	8.5±1.5	<0.001**
Monthly income (U.S. Dollar); median (range)	93 (31 to 1,562)	781 (93 to 4,687)	<0.001**
Occupation; n (%)			<0.001**
Housework	1 (2.5)	3 (7.9)	
Self-employed	9 (22.5)	11 (28.9)	
Merchant	1 (2.5)	3 (7.9)	
Government officer	0 (0.0)	17 (44.7)	
Company employee	0 (0.0)	2 (5.0)	
Personal business	0 (0.0)	2 (5.0)	
Agriculture	29 (72.5)	0 (0.0)	
Education; n (%)			<0.001**
Primary school	32 (80.0)	7 (18.4)	
Middle school	4 (10.0)	4 (10.5)	
High school	3 (7.5)	6 (15.8)	
Bachelor degree	1 (2.5)	14 (36.8)	
Higher Bachelor degree	0 (0.0)	7 (18.4)	
Sunscreen usage; n (%)			0.010**
Not use	24 (60.0)	12 (31.6)	
Use	16 (40.0)	26 (68.4)	

BMI=body mass index; SD=standard deviation

* Student's t-test, ** Significant p<0.05

Table 2. Baseline chemistry levels of participants (outdoor group vs. indoor group) (n=78)

Baseline laboratory	Outdoors (n=40); mean±SD	Indoors (n=38); mean±SD	p-value*
BUN (mg/dL)	12.4±4.0	11.7±2.8	0.380
Creatinine (mg/dL)	0.7±0.1	0.7±0.1	0.750
AST (U/L)	22.5±5.9	21.2±4.8	0.300
ALT (U/L)	17.4±5.8	17.2±7.8	0.910
Albumin (g/dL)	4.1±0.2	4.3±0.32	0.110

BUN=blood urea nitrogen; AST=aspartate aminotransferase; ALT=alanine aminotransferase; SD=standard deviation

* Student's t-test

the outdoor group and 77.5% in the indoor group. Although, participants that worked indoors had a higher risk of vitamin D insufficiency than those worked outdoor, it was without a statistical difference (RR 2.00, 95% CI 0.71 to 5.33). When vitamin D deficiency was defined as 25(OH)D level <20 ng/mL, the prevalence of vitamin D deficiency were 12.5% and 25.0%, in the outdoor group, and indoor group, respectively, with no statistical significance (RR 1.35, 95% CI 0.98 to 1.85).

The serum calcium levels in both groups were in a normal range. When calcium level was corrected by albumin, the serum levels in the outdoor group and the indoor groups were significantly different (p=0.005). Corrected serum calcium was 9.2±0.4 mg/dL and 9.0±0.3 mg/dL in the outdoor group and indoor group, respectively. The data is presented in Table 3.

Table 3. Serum 25(OH)D and serum calcium levels in outdoor group vs. indoor group

Chemistry laboratory	Outdoors (n=40); mean±SD	Indoors (n=38); mean±SD	p-value*
25(OH)D (ng/mL)	29.3±8.6	26.4±8.6	0.130
Calcium (mg/dL)	9.4±0.4	9.2±0.4	0.070
Corrected calcium (mg/dL)	9.2±0.4	9.0±0.3	0.005**

SD=standard deviation

* Student's t-test, ** Significant p<0.05

Table 4. Subgroup analysis of serum 25(OH)D and serum calcium in perimenopausal and postmenopausal women

Group	Menopausal stage	n	25(OH)D (ng/mL); median (range)	Calcium (mg/dL); median (range)	Corrected calcium (mg/dL); median (range)
Outdoor	Perimenopause	12	30.4 (20.9 to 49.6)	9.2 (8.7 to 10.0)	9.07 (8.7 to 9.7)
	Postmenopause	28	29.1 (11.2 to 45.5)	9.4 (8.3 to 10.5)	9.3 (8.0 to 10.0)
p-value*			0.342	0.373	0.358
Indoor	Perimenopause	29	23.4 (13.9 to 34.8)	9.0 (8.7 to 9.9)	8.6 (8.5 to 9.5)
	Postmenopause	9	26.4 (11.6 to 52.9)	9.2 (8.6 to 10.4)	9.0 (8.4 to 10.0)
p-value*			0.566	0.840	0.81

* Mann-Whitney U test

Table 5. Comparison of vitamin D insufficiency [25(OH)D <30 ng/mL] by RR between outdoor and indoor groups

Group	<30 ng/mL; n (%)	≥30 ng/mL; n (%)	RR	95% CI*
Outdoor	23 (57.5)	17 (42.5)	2.00	0.71 to 5.33
Indoor	31 (77.5)	9 (22.5)		

RR= relative risk; CI=confidence interval

* Chi-square test

Table 6. Comparison of vitamin D deficiency [25(OH)D <20 ng/mL] by RR between outdoor and indoor group

Group	<20 ng/mL; n (%)	≥20 ng/mL; n (%)	RR	95% CI*
Outdoor	5 (12.5)	35 (87.5)	1.35	0.98 to 1.85
Indoor	10 (25.0)	30 (75.0)		

RR= relative risk; CI=confidence interval

* Chi-square test

The subgroup analysis of the perimenopausal and postmenopausal group found that 25(OH)D, calcium, and corrected calcium were not significantly different between the two groups. The data is shown in Table 4.

The rate of vitamin D insufficiency and deficiency by relative risk between the outdoor group and the indoor group were comparable as shown in Table 5 and 6, respectively.

Participants in the present study who used sunscreen had lower 25(OH)D levels than those who did not at 26.3±6.4 ng/mL and 29.8±10.6 ng/mL, without statistical significance (p=0.090). Moreover, the indoor group had a higher rate of using sunscreen than the outdoor group with statistical significance.

Discussion

Serum 25(OH)D (calcidiol) is easy to measure and is an appropriate parameter to detect vitamin D in the human body. Nevertheless, there is no certain rate used as an official criterion of vitamin D serum level measurements to verify vitamin D deficiency in Thailand. Nowadays, routine screening for serum vitamin D levels is not recommended for routine clinical practice due to the high cost. Moreover, the test is not available in every hospital. In addition, the results of vitamin D levels depends on equipment, which may vary in terms of measurement unit such as nmol/mL and ng/mL.

A study in Thailand regarding the prevalence of vitamin D insufficiency in Thai premenopausal women revealed that 77.85% in the rural area in one of the Northeast provinces had a mean serum 25(OH)D level of 29.09 ng/mL[18]. On the contrary, the prevalence of vitamin D insufficiency in the selected elderly Thai women in the rural area was only 17.9%[16]. The high prevalence of vitamin D insufficiency in the first paper might be from the cut off level of insufficiency with 35 ng/mL instead of 30 ng/mL. While the low percentage in the second paper might be explained by the lifestyle of people in the rural area that stay at their own home and not a nursing home, and they might be in good health and did not stay in the shade, most of the time but go out to visit with their nearby neighbors, with a greater chance for sun exposure. Moreover, the older population in the rural area do not routinely use sunscreen.

Another study in Thai people including adults of both genders, reveals the highest rate of vitamin D insufficiency in Bangkok at 64.6% while the percentage of vitamin D insufficiency in other parts were 46.7% and 33.5% in the municipal areas and other parts of Thailand, respectively. In the present study, one of the risk factors for vitamin D insufficiency except for the female gender and living in Bangkok was the young age population[7]. This phenomenon was also found in Korea[19]. That may be from lifestyle changes including diet, physical activity with sun exposure avoidance, and widespread routine use of sunscreen protection. Therefore, the topic of young adults and the high risk of vitamin D insufficiency should be of concern[20].

From the present study, serum vitamin D levels in perimenopausal and postmenopausal women in Chiang Mai were low with the mean of 25(OH)D 27.9±8.7 ng/mL. The prevalence of vitamin D insufficiency in the outdoor group and indoor group was 57.5% and 77.5%, respectively, while the prevalence of vitamin D deficiency was 12.5% and 25.0%, respectively. The mean serum 25(OH)D levels of the outdoor group was 29.3±8.6 ng/mL and was slightly higher than the indoor group, which was 26.4±8.6 ng/mL, without statistical significance.

Levels of serum calcium were related to vitamin D levels as they were found higher in the outdoor workers. Despite low serum vitamin D levels, the calcium levels remained normal. When corrected by albumin, the corrected calcium was significantly higher in the outdoor workers than the indoor workers. The difference in corrected calcium was explained by the different albumin levels that were higher in the indoor group.

The baseline demographic data in terms of monthly income was significantly lower in the outdoor group reflecting a lower level of education. Most of them choose their career with agriculture and none work as a government officer or company employee.

Data regarding occupation, income, education, and rate of sunscreen usage, implied that the outdoor workers had a lower socioeconomic status than that of the indoor workers. It was assumed that in terms of nutrition, the indoor workers might have better nutrition, including higher calcium and vitamin D levels, which was a factor that caused the comparison of vitamin D levels between the indoor and the outdoor workers to be not significantly different.

However, the indoor workers may still have a chance of getting sunlight regularly such as on the way to work and to have lunch. Thailand is a sunny, tropical country with 2.0 to 9.1 hours per day[7]. Moreover, one study showed that indoor workers were exposed to different light sources such as fluorescent light bulbs, computers, televisions, and tablets that emitted UVB wavelength less than 280 nm, and enabled vitamin D synthesis in the skin[21]. On the other hand, the outdoor workers used various items to protect them from sunlight such as hats, facial masks, shawls, long sleeve shirts, and umbrellas.

From the present study, both the outdoor and indoor workers applied sunscreen to protect from UV rays that might affect vitamin D synthesis. Subsequently, working outdoors or indoors could not be used as the criterion to determine vitamin D distribution in menopausal women. In the further study, other aspects such as sunscreen usage, clothes protection, and dietary intake should explore the correlation with vitamin D level.

The limitation of the present study was the low number of participants. Moreover, data such as food intake including diary product are lacking. Further study with larger numbers of perimenopausal and postmenopausal women working outdoors and indoors, and detail of food intake should be done to compare and confirm the difference in serum vitamin D level.

Conclusion

The mean serum 25(OH)D level in perimenopausal and postmenopausal women in Chiang Mai was low. In addition, the serum 25(OH)D level of the participants who worked outdoors and indoors was not different. Hence, sunlight exposure cannot be the reliable factor to ensure the promising adequate levels of vitamin D. Therefore, a high vitamin D diet or vitamin D supplement should be recommended for all perimenopausal and postmenopausal women. Furthermore, vitamin D supplements should be widely distributed to all hospitals and pharmacies. It is very important to provide knowledge of the consequence of vitamin D deficiency and sources of vitamin D including vitamin D supplements to the public.

What is already known on this topic?

The authors have already known that menopausal women and the elderly are the high-risk group for vitamin D insufficiency and deficiency. Even though Thailand is a sunny country for the whole year, there is a high rate of vitamin D deficiency. Inadequate sunlight exposure leads to vitamin D deficiency and data have shown this relationship. However, limited data in the selected group of menopausal women has been reported.

What this study adds?

This study was conducted on peri- and post-menopausal women to explore and compare the level of serum vitamin D between the women who work outdoors and indoors, which was not different. The present study is providing valuable information. Based on this, we are recommending a high vitamin D diet or vitamin D supplement to all perimenopausal and postmenopausal women.

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Conflicts of interest

The authors declare no conflict of interest.

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