Frequency and Determinants of Vitamin D Deficiency Among Premenopausal and Postmenopausal Women: A Multicenter Study in Karachi, Pakistan

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Abstract

Background: Vitamin D deficiency is becoming a serious public health problem, even in sun-drenched cities like Karachi, Pakistan. We investigated the prevalence of vitamin D deficiency VDD and its association with sociodemographic characteristics, anthropometric measures, and lifestyle factors among Pakistani women (n = 784).

Methods: Face-to-face interviews were conducted to collect information from premenopausal and menopausal women and serum 25(OH)D was measured after the interview.

Results: The mean age of the women was 46.2 years, and the mean 25-hydroxyvitamin D (+/- SD) levels were 22.9 ng/ml (+/-20.3). A total of 57% of women were vitamin D deficient with higher vitamin D deficiency found among premenopausal (64.7%) women compared to menopausal women (49%). Factors associated with vitamin D deficiency were lower socioeconomic status (OR 2.00; 95% CI 1.15–3.48), younger age with highest vitamin D deficiency in < 35 years of age (OR 3.11; 95% CI 1.76–5.51), and winter season (OR 1.51, 95% CI 1.07-2.15) after adjusting for multiple confounders. The use of vitamin D supplement use (OR 0.59, 95% CI 0.38-0.92) and vigorous exercise (OR 0.20, 95 % CI 0.05-0.80) was protective against vitamin D deficiency.

Conclusions: The study shows a high prevalence of vitamin D deficiency, with detrimental health effects among younger women belonging to lower socioeconomic status during the winter season. The use of vitamin D supplements and vigorous exercise were protective measures. Public health campaigns are needed for education and awareness about vitamin D deficiency to improve vitamin D status for younger women living in poor environments.

Background

Vitamin D deficiency VDD (< 20 ng/ml) is becoming common and increasing in majority of population worldwide. There is also a lot of research interest in studies assessing the role of VDD in many diseases including the recent pandemic of Covid 19(1, 2). VDD is an important public health problem in Pakistan too. According to a study in Hyderabad, both VDD and vitamin D insufficiency was 78.3% among 1244 healthy individuals (3). In another study, at Ayub Teaching Hospital Abbottabad, high VDD was reported among 202 patients (63.4%) with complaints of generalized body aches (4). A study conducted among healthy women of child-bearing age in Lahore reported that VDD was 73 %. Factors associated with VDD were lesser sun exposure, illiteracy, winter season during sampling and no usage of multivitamins were the risk factors. A cross-sectional population survey among 300 adult population of Karachi showed median level of serum vitamin D as 18.8 ng/dL with high prevalence of VDD (5). VDD was reported as 90.1% among 305 premenopausal females in a cross-sectional study in Karachi, Pakistan. Factors associated with VDD were age, city of residence in downtown and suburbs, and type of housing (6). In a recent study among 221 medical students of a medical school, VDD was found in 89.14 % and factors associated with VDD were limited sun exposure and attire consisting of full length of sleeves outside (7).

There is a lot of variation in the vitamin D status of women living in Karachi in different studies, most of which consisted of smaller sample sizes and only few factors causing VDD were assessed. Therefore, the objective of the study was to investigate all the factors associated with VDD among women of both premenopausal and menopausal status in Karachi Pakistan.

Methods

Study Population

We extracted data of 784 women, who were in the control group and had complete information on potential predictors of VDD and serum 25(OH)D concentrations, from the case control study (the details of which are mentioned in a recently published article) (8). All the women were interviewed attending in- and out-patient services for general medical, and surgical departments. Exclusion criteria were history of any cancer, parathyroid, renal or liver disease, chronic malabsorption syndromes or any known or suspected drug that may alter vitamin D metabolism.

Face to face interviews were conducted and information was collected about sociodemographic factors, clinical and reproductive history, sun exposure, use of vitamin D supplementation, and anthropometric measurement. Age was categorized into < 35, 35–45, 46–54, > 55 years. Education was categorized as < 8, 8–12, or > 12 years of education. Socio-economic (SES) factors included were multiple variables like crowding index (the number of household members divided by number of rooms) and was further categorized as < 1, 1–2, > 2. A composite variable was calculated for socioeconomic status by factor analysis of important variables like education, place and type of residence, crowding index, home ownership, number of rooms, total household members, total household monthly income. Finally, SES was categorized into upper, middle and lower status (8).

Sun exposure measurement questionnaire

The validated Long-Term Sun Exposure Measurement Questionnaire (LT SEM-Q) was used to assess sun exposure among women (9). This questionnaire assessed sunlight exposure, duration of sun exposure, skin tone, use of sunscreens and sun avoidance behaviour. The time spent outdoors between 10 am and 4 pm was asked to calculate approximate amount of time in minutes per week women were exposed to UVB radiations. Details of weights given to sun exposure variables are in the supplementary table A. The final scoring algorithm of sun exposure score in summers and winters was created by multiplying the time (minutes) spent in the sun by the proportions of the different variables. The skin tone of the women was assessed using a shade card (10 skin tones (1–10)) by matching shade of the skin on forehead with the shade on the card, according to LT SEM-Q.

Vitamin D supplements

Use of vitamin intake was asked in detail as types of intake, duration and form of intake as injectable, oral drops or capsules apart from intake as yes vs. no.

Measurement of serum 25(OH)D concentrations

Venous blood samples (2.5 ml) were taken from all participants and collected in yellow topped gel tubes at the end of the interview. Serum 25(OH)D concentrations were measured by a kit from DiaMetra S.r.l. Headquarter: Via Garibaldi, 18-20090 SEGRATE (MI) Italy, using solid phase enzyme-linked immunoassay (ELISA). The cut-off used to define VDD was according to the Parathyroid Hormone (PTH) levels (10).Vitamin D deficiency was defined as a serum 25-hydroxyvitamin D concentrations ≤ 20 ng/ml. Season of blood collection (winter/ spring vs. summer /fall) was also recorded.

Other variables included fish consumption (asked as no intake vs. least once per month vs. more) and BMI (calculated as weight in kilograms divided by the square of height in meters. Women were categorized for their BMI according to the World Health Organization (WHO) cut off for BMI (11): a normal weight (18.5–24.9 kg/m2), being overweight (25.0–29.9 kg/m2) or obese (> 30 kg/m2).

Statistical analysis

Mean and standard deviations are reported for continuous variables and frequencies and percentages for categorical variables to illustrate the differences in vitamin D concentrations between premenopausal and menopausal women. The crude and adjusted ORs and 95 % CI s were calculated for VDD (vitamin D level lower than 20 ng/ml versus a non deficient vitamin D status) through logistic regressions while entering the following variables as predictors in the model: age, education level, parity, SES, season, BMI, use of vitamin D supplements, fish consumption, exposure to sunlight, sun avoidance behaviors.

Statistical analysis of the data was carried out using the SPSS package for Windows 22.0 (SPSS, IBM, Armonk, NY, USA).

Results

Mean 25(OH)D level was 22.9 ng/ml (SD 20.3) and median level was 16.7ng/ml. There was high prevalence of VDD among both pre and postmenopausal women. Individual 25(OH)D concentrations ranged from 0.3 ng/ml to 165.5 ng/ml. The median 25(OH)D level was lower among premenopausal women (13.9 ng/ml) compared to menopausal women (20.4 ng/ml).

Women were further categorized into four different concentrations of 25(OH)D, defined as severely deficient (< 12 ng/ml), deficient (12–19 ng/ml), insufficient (20-30ng/ml) and sufficient (> 30ng/ml) (Table 1). Both severe vitamin D deficiency and deficiency was significantly more frequent in premenopausal women compared to menopausal women.

Table 1
Number and percentage of women for different serum 25(OH)D concentrations

Serum 25(OH)D (ng/ml)		Premenopausal women		Menopausal women		All women		p value*
		n	%	n	%	n	%	
								< 0.001
Severely deficient	**< 12**	124	39.9%	78	26.2%	202	33.2%	
Deficient	**12–19**	77	24.8%	68	22.8%	145	23.8%	
Insufficient	**20–30**	52	16.7%	61	20.5%	113	18.6%	
Sufficient	**> 30**	58	18.6%	91	30.5%	149	24.5%	

**p values generated from Chi-square or Fisher Exact test*

Table 2 shows that VDD was higher in younger compared to older women. The majority of the women had skin tone of 5 and 6 categorized as wheatish tone, however, skin tone was not associated with serum 25(OH)D levels. As it was a hospital-based study, it was observed that a higher percentage (63%) of women took vitamin D supplements and fish intake was less frequent and nearly 40 % women ate no fish at all.

Table 2
Socio-demographic and clinical characteristics of both premenopausal and menopausal women (n = 784) with and without vitamin D deficiency in Karachi, Pakistan

Categories	vitamin D > 20		Vitamin D deficiency ≤ 20		OR (95% CI)
	n	%	n	%	
Age groups					
< 35	29	10.9	65	18.7	3.46 (2.00, 5.97)
35–45	77	28.9	144	41.4	2.88 (1.88, 4.43)
46–54	69	25.9	80	23.0	1.79 (1.13, 2.83)
55 & above	91	34.2	59	17.0	Ref
Education					
<grade8	49	18.4	68	19.5	1.07 (0.69, 1.65)
grades 8–12	87	32.7	111	31.9	0.98 (0.68, 1.41)
>grade12	130	48.9	169	48.6	Ref
Socioeconomic status					
upper	61	22.9	52	14.9	2.62 (1.58, 4.34)
middle	158	59.4	191	54.9	1.42 (0.92, 2.17)
lower	47	17.7	105	30.2	Ref
Parity [a]					
nullipara	34	12.8	59	17.0	0.82 (0.50, 1.350
43833	136	51.1	152	43.7	0.64 (0.40, 1.04)
> 3	96	36.1	137	39.4	Ref
menopause					
menopause	152	58.0	146	43.0	1.90 (1.38, 2.63)
premenopause	110	42.0	201	57.0	Ref

[c]Parity was restricted to women who ever had a full-term pregnancy (a pregnancy was considered as full-term if it resulted in a live birth or lasted 7 or more months)

[b]BMI, body mass index; BMI was categorized according to the WHO classification for Asian as underweight/normal weight (< 23 kg/m2), overweight (23–25 kg/m2) or obese (_26 kg/m2).

Abbreviations: OR, odds ratio CI, confidence interval

Reference: OR are compared to those women who had VDD

Categories	vitamin D > 20		Vitamin D deficiency ≤ 20		OR (95% CI)
history of any comorbid					
yes	161	60.5	168	48.3	0.61 (0.44, 0.84)
no	105	39.5	180	51.7	Ref
season vitamin D					
Winter	**102**	39.2	167	48.7	1.47 (1.06, 2.04)
summer	**158**	60.8	176	51.3	Ref
fish consumption					
no intake	107	41.2	136	40.1	1.14 (0.56, 2.29)
<than once a month	136	52.3	184	54.3	1.21 (0.61, 2.42)
> once /month	17	6.5	19	5.6	Ref
Vitamin D supplements intake					
Yes	204	76.7	172	49.4	0.30 (0.19, 0.46)
No	62	23.3	176	50.6	Ref
Body mass index [b]	28.03	5.2	27.94	5.06	1.00 (0.97, 1.03)

[c]Parity was restricted to women who ever had a full-term pregnancy (a pregnancy was considered as full-term if it resulted in a live birth or lasted 7 or more months)

[b]BMI, body mass index; BMI was categorized according to the WHO classification for Asian as underweight/normal weight (< 23 kg/m2), overweight (23–25 kg/m2) or obese (_26 kg/m2).

Abbreviations: OR, odds ratio CI, confidence interval

Reference: OR are compared to those women who had VDD

Table 3 shows lifestyle and sun exposure factors related to VDD.

Table 3
Lifestyle and sun exposure related factors associated with serum 25-hydroxyvitamin in Pakistani women.

Categories	**vitamin D ≥ 20**		**Vitamin D deficiency < 20**		
Vigorous exercise	**n**	**%**	**n**	**%**	**OR (95% CI)**
< 3.5hrs/wk	17	6.4	7	2.0	0.82(0.29, 2.28)
> 3.5 hrs/wk	6	2.3	10	2.9	0.25 (0.07, 0.95)
no exercise	243	91.4	331	95.1	
Moderate Exercise					Ref
< 3.5hrs/wk	32	12.0	58	16.7	0.59 (0.40, 0.87)
> 3.5 hrs/wk	54	20.3	98	28.2	1.00 (0.58, 1.72)
no exercise	180	67.7	192	55.2	Ref
Walk					
< 3.5hrs/wk	78	29.3	88	25.3	1.59(1.00, 2.53)
> 3.5 hrs/wk	46	17.3	44	12.6	1.18 (0.71, 1.97)
no exercise	142	53.4	216	62.1	Ref
Head Covered					
Yes	171	64.3	271	78.1	1.98 (1.39, 2.83)
No	95	35.7	76	21.9	Ref
Face Covered					
Yes	39	14.7	83	23.9	1.83 (1.20, 2.79)
No	227	85.3	264	76.1	Ref
Neck Covered					
Yes	129	48.5	208	59.9	1.59 (1.15, 2.19)
No	137	51.5	139	40.1	Ref
Full Arm Covered					
Yes	166	62.4	255	73.5	1.67 (1.18, 2.36)
No	100	37.6	92	26.5	Ref

Abbreviations: OR, odds ratio CI, confidence interval
Reference: OR are compared to those women who had VDD

Categories	vitamin D ≥ 20		Vitamin D deficiency < 20		
Half Arm Covered					
Yes	100	37.6	88	25.4	0.56(0.40,0.80)
No	166	62.4	259	74.6	Ref
Attire outside					
chadder	70	26.3	91	26.3	1.44(0.95, 2.19)
burqa	92	34.6	161	46.5	1.94(1.33, 2.83)
others	104	39.1	94	27.2	Ref
Skin Tone					
Dark	151	59.0	198	59.1	1.01(0.72, 1.42)
Wheatish	93	36.3	123	36.7	0.89 (0.40, 1.98)
Fair	12	4.7	14	4.2	Ref
Season of blood draw					
winter	158	60.8	176	51.3	1.47 (1.06, 2.04)
summer	102	39.2	167	48.7	Ref
Sun avoidance behavior					
Yes	215	80.8	296	85.3	1.38 (0.90, 2.11)
No	51	19.2	51	14.7	Ref
Sunlight exposure score (summer)*	26.47	75.6	32.89	104.7	10.01(0.97, 10.06)
Sunlight exposure score (winter)*	39.96	89.6	49.08	121.1	10.02 (9.98, 10.05)
Abbreviations: OR, odds ratio CI, confidence interval					
Reference: OR are compared to those women who had VDD					

Table 4 shows the factors associated with vitamin D deficiency were lower socioeconomic status (adjusted OR 2.00; 95% CI 1.15–3.48), younger age with highest vitamin D deficiency in < 35 years of age (adjusted OR 3.11; 95% CI 1.76–5.51) and winter season (adjusted OR 1.51, 95% CI 1.07-2.15) after adjusting for multiple confounders (p<0.0001). Use of vitamin D supplement use (adjusted OR 0.59, 95% CI 0.38-0.92) and vigorous exercise (adjusted OR 0.20, 95 % CI 0.05-0.80) were protective against vitamin D deficiency.

Table 4
Final table showing adjusted ORs (95% CI) of factors associated with vitamin D deficiency among women in Karachi Pakistan.

Variables	Adjusted OR	95% CI	p value
Age			< 0.001
< 35	3.11	1.76, 5.51	
35–45	2.66	1.69, 4.19	
46–54	1.72	1.07, 2.77	
55 & above	Ref	.	.
Socioeconomic Status			0.029
Lower	2	1.15, 3.48	
Middle	1.23	0.77, 1.95	
Upper	Ref	.	
Season of blood withdraw			0.05
Winter	1.51	1.07, 2.15	
Summer	Ref		
Vitamin D supplementation use			<0.001
Yes	0.59	0.38, 0.92	
No	Ref		
Vigorous exercise			0.027
< 3.5 hrs/wk	0.63	0.21, 1.86	
> 3.5 hrs/wk	0.2	0.05, 0.80	
No exercise	Ref		

Adjusted for education, BMI, parity, skin tone, attire outside. Reference: OR are compared to those women who had no vitamin D deficiency. Abbreviations: OR odds ratio, 95 CI = 95 % confidence interval

Discussion

In our study, 57% of women had VDD and only 24.5% of participants had optimal vitamin D status. Risk of VDD was associated with younger age, lower SES, winter season, while exercise and vitamin D supplement use were protective. A cross-sectional study among 351 patients at the out-patient department of General Medicine of a hospital in Islamabad, reported VDD present in 62.9% of females (mean age 46.03 +/- 16.18 years) and the mean vitamin D level was 14.09 +/- 12.93 ng/ml (12). In South Florida (U.S.), the mean (+/-SD) of 25(OH)D level was 22.4 +/- 8.2 ng/ml in 40% of women during winter(13). A study conducted in Lahore similarly reported high prevalence of Vitamin D deficiency among healthy women of child-bearing and was also associated with low education and lack of proper sun exposure and multivitamin intake (14).

In a study conducted at an urban hospital in Boston, VDD was associated with winter season, higher body mass index, and physical inactivity(15). In a study in Japan among 4,793 patients with rheumatoid arthritis, the mean (SD) serum 25(OH)D level was 16.9 ng/mL (6.1), and the prevalence of vitamin D deficiency was 71.8%. Predictors of VDD were female gender, younger age, among other factors that included low serum levels of total protein and total cholesterol, high serum ALP levels, and NSAID (16). VDD was also found high among pregnant women in Belgium (17). Similar to our study finding of association of VDD with younger age, vitamin VDD was found higher in younger women on other studies too (18-22).

Similar to our study, VDD is found to be more common among women during winter/spring compared with summer/autumn (23-25). A study in 3,327 pregnant Japanese women, VDD prevalence was 73.2% and it was higher in April after the winter season. Sun exposure of >15 mins for 1-2 days / week and usage of dietary vitamin D were protective against VDD (26).

A study conducted in Jinnah Postgraduate Medical Center, Karachi among students showed that VDD was more common in winter (27). A cross-sectional study of 14,302 Chinese participants aged 18-65 years from six major cities in China reported VDD was higher among females, in spring and winter from certain residential regions (28).

A study conducted among pregnant women in Malaysia reported that intake of vitamin D was protective against VDD. There was no association of VDD observed with age, educational, SES, employment, parity, body mass index, sun exposure(29). Another small cross-sectional study in Riyadh, Saudi Arabia, reported VDD among 60.2% of participants was associated with lack of usage of vitamin D, multi-vitamins (30). Another study in Riyadh, Saudi Arabia reported that absence of vitamin D supplements usage, younger age were factors associated with VDD among females(31). VDD was 75.1% in a study in France and was associated with no intake of vitamin D (32). A cross-sectional study of 634 healthy volunteers aged 18-50 years reported VDD associated with lack of multivitamin use ($P<0.001$ for each predictor)(33).

Our study showing lower SES and sedentary lifestyle associated with VDD was also reported by The National Health and Nutrition Examination Survey (NHANES) 2001-2010 (34). A cross-sectional study in Saudi Arabia, reported younger age, less exercise, less Vitamin D intake, as predictors of VDD (35).

Our study results are consistent with findings in few other studies conducted in Switzerland, China and New South Wales showing association of VDD with childbearing age, winter season and no usage of vitamin D supplement (36-38). A large study from the Korean National Health and Nutrition Examination

Survey (KNHANES) 2008-2009 among 2062 adolescents also showed that VDD was higher in senior high school students and was associated with winter season and parental vitamin D deficiency (39).

Contrary to previous studies across different ethnic backgrounds, this study within Pakistani females showed that skin tone, obesity, etc were not associated with VDD. There was no association observed between VDD and sun exposure questionnaire. In a study in Brazil, the use of sun exposure questionnaire with 25OHD level showed low accuracy and its lack of discrimination between vitamin D sufficient and deficient individuals (40). A cross-sectional study among 254 university students in Shiraz also showed no association between VDD and sun exposure(41). Moreover, there was no association found between dietary sources of vitamin D and VDD. One reason could be the low consumption of fish and milk and vitamin D fortified food items in our population.

Our study has several strengths. The study collected comprehensive information on all potential factors associated with VDD. The data was collected by trained medical officers with expertise in both medical history taking and research. The main weakness of the study is the recall bias which is inherent in any cross sectional or case control study. Awareness programs and education about the importance of adequate vitamin D levels, are needed for sensible sunlight exposure and adequate nutritional intake of vitamin D-rich foods to prevent adverse health outcomes related to vitamin D deficiency. Supplement use of Vitamin D is particularly important especially during the current pandemic of Covid 19.

Abbreviations

CI: Confidence Interval, VDD: Vitamin D deficiency, OR: Odds Ratio; AKU: Aga Khan University.

Declarations

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Availability of data and materials The datasets used and analyzed during the current study are available from the corresponding author on request.

Ethics approval and consent to participate

The study was performed in accordance with the ethical standards of the Declaration of Helsinki (1964) and its subsequent amendments. The ethical approval was obtained by the Human Research Ethics Committee of the University of Adelaide, Australia (H-2014-111) and the Ethical Review Committees of Aga Khan University Hospital, Karachi Pakistan (3074-CHS-ERC-14). Informed consent was obtained using the Patient Information Sheet and Consent form. Participants were clearly informed about the objectives and procedures of the study, their rights and commitments, and the benefits and risks involved. If the patients agreed to participate, they were asked to sign the consent paper. The consent form of the study was provided both in English and the local language of Urdu. If unable to read the consent form, the form was read out to participants and informed consent was taken verbally.

Consent for publication Not applicable.

Competing interests The authors declare that they have no conflict of interest.

Authors' contributions US participated in the design of the study, acquisition of data, performed the statistical analysis, and drafted the manuscript. DC made substantial contributions to conception and design, interpretation of data, acquisition of data, and interpretation of data. IA performed the statistical analysis and contributed to interpretation of data. AS & DS made contribution to manuscript writing and proofreading it. All authors were involved in revising the manuscript critically for important intellectual content, and final approval of the version to be published.

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