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Diabetes & Metabolic Syndrome: Clinical Research & Reviews 13 (2019) 1517–1521

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Original Article

Vitamin D status, determinants and relationship with biochemical profile in women with Type 2 Diabetes Mellitus in Delhi, India

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ARTICLE INFO

Article history:
Received 3 February 2019
Accepted 5 March 2019

Keywords:
Vit D deficiency
Diabetes
Diabetes
Women's health
India

ABSTRACT

Objective: To determine the burden of vitamin D deficiency and its determinants and to assess the relationship of 25 hydroxycholecalciferol (25-OHD) levels with biochemical parameters linked to health outcomes in women with Type 2 Diabetes Mellitus (T2DM).

Material and methods: This was a hospital based cross-sectional study in the diabetes out-patient department clinic of a major tertiary care hospital in Delhi, India. Adult women with T2DM on treatment for at least 6 months were included in this study. The women who have been given Vitamin D supplementation during the past 6 months were excluded. We assessed Serum 25-OHD, HbA1c, lipid profile and fasting plasma glucose in the patients through standardized laboratory methods.

Results: One hundred women with T2DM were enrolled of which 22 (22%) had good glycemic control (HbA1c $< 7\%$). Vitamin D deficiency was seen among 77 (77%) and insufficiency among 16 (16%) of the recruited subjects. Younger age group (31–45 years) and illiteracy was significantly associated with vitamin D deficiency ($p < 0.05$). No association was found between Vitamin D deficiency and HbA1c levels.

Conclusion: Vitamin D deficiency is highly prevalent among women with T2DM. Illiteracy and young age were major determinants of vitamin D deficiency indicating they need special attention and Vitamin D supplementation.

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1. Introduction

Non Communicable diseases (NCDs) impact women's health across their lifecycle. NCDs account for 65% of all female deaths worldwide and are a significant cause of female morbidity and mortality during their childbearing years [1]. Vitamin D deficiency has been reported as one of the risk factors in the development of chronic diseases like Type 2 Diabetes Mellitus (T2DM), cardiovascular diseases and autoimmune diseases. Vitamin D has been suggested to influence metabolic pathways relating to beta cell function although the mechanisms underlying the same are incomplete understood [2,3].

The reported prevalence of vitamin D deficiency or insufficiency in patients with T2DM varies from 70 to 90% [4–8]. A cross-sectional study in South India observed Vitamin D deficiency in 84% T2DM patients [9]. Women can be at higher risk of Vitamin D deficiency in the developing world due to poor dietary practices and lack of sun exposure due to conservative cultural practices like full-bodied clothing, cosmetic concerns and increased time spent indoors [10].

Furthermore, Vitamin D is suspected to influence biochemical parameters that are linked to health outcomes in Diabetes patients. A few studies have linked Vitamin D deficiency with impairment of glycemic control [11,12]. Low Vitamin D intake may lower HDL [13] while Vitamin D supplementation can also lower blood cholesterol levels [14].

Since, Vitamin D deficiency can be corrected by dietary modifications and nutritional supplementation; ascertaining the

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https://doi.org/10.1016/j.dsx.2019.03.005
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relationship between Vitamin D deficiency and biochemical parameters in T2DM patients is important.

This study was therefore conducted with the objective of determining the burden of vitamin D deficiency and its determinants and to assess the relationship of 25 hydroxycholecalciferol (25-OHD) levels with biochemical parameters (Total Cholesterol, TGs, LDL, HDL, HbA1c) linked to health outcomes in women with T2DM.

2. Methods

This was a hospital based cross-sectional study conducted in the Diabetes out-patient department (OPD) clinic of a major tertiary care hospital in Delhi, India. Data was collected from January 2017 to December 2017. Adult women with T2DM on treatment for at least 6 months were included in this study. The women who have been given Vitamin D supplementation during the past 6 months were excluded.

The study subjects were recruited from among the women with Type 2 Diabetes Mellitus attending the diabetes clinic OPD sessions by simple random sampling using a lottery method. A total of 10 women were selected on each clinic day for a larger study to assess their diabetes self-care practices, of which 3 women were further selected by lottery method for estimation of their Vitamin D levels. A total of 100 women were evaluated for Vitamin D levels for this study.

A pre-designed, pre-tested, semi-structured interview schedule was used for data collection. Clinical parameters like type of anti-diabetes therapy and presence of co-morbidities were validated using the patient medical records. Dietary history was taken by recall method for previous seven days from the day of interview. The socio-economic status (SES) of the study subjects was ascertained using Modified BG Prasad Scale adjusted for the current consumer price index (CPI–W, 2017) [15].

A fasting venous sample of 7 ml was collected using aseptic precautions by venipuncture. For estimation of HbA1C 1 mL of blood was added to ethylenediaminetetraacetic acid (EDTA) vial, five mL to plain vial for estimation of lipid profile and Vitamin D levels and one mL of blood was added to sodium fluoride (NaF) vial for the estimation of fasting plasma glucose.

Vitamin D deficiency was defined as Serum 25(OH) D levels less than 20 ng/ml, insufficiency as levels less than 30 ng/ml and sufficiency as levels of 30 ng/ml or above. Further Vitamin D deficiency was sub classified as: Mild: if 25(OH)D level was 10–20 ng/ml, moderate if it was 5–10 ng/ml and severe if was <5 ng/ml. This classification is validated as per the electrochemiluminscence based immunoassay method [16].

Based on the American Diabetes Association (ADA) Diabetes Management Guidelines 2016 and ICMR Guidelines for Management of Type 2 Diabetes, good glycemic control was defined as HbA1c level of <7% [17]. Glycated Hemoglobin (HbA1c) was assayed by HPLC method by using reagent from Bio-Rad Laboratories Inc. (UK) adopted to D10 fully automated HPLC based analyser (Bio-Rad, UK). Fasting Plasma Glucose level was estimated using commercially available GOD-POD based glucose assay kit from Randox Pvt Ltd. (UK) adopted to AU480 fully automated random access clinical chemistry analyser (Beckman Coulter Inc., USA).

Lipid Profile estimation: The lipid parameters e.g. Triacylglycerol total cholesterol, LDL-C and HDL-C were assayed from serum samples by specific enzymatic methods by using commercially available reagent kit from Randox Laboratories Ltd (London U.K) adapted to Beckman Coulter AU 400 clinical chemistry analyser.

Statistical Analysis: The collected data was entered into MS-EXCEL, cleaned and then analyzed using SPSS-PC-22 version (SPSS Inc. USA). Qualitative data was expressed in frequency and proportions and quantitative data as mean and standard deviation. The difference in proportions was assessed applying the Chi-square or Fisher's exact test and difference in means between groups by the independent samples *t*-test. A P value < 0.05 was considered as statistically significant.

Ethical Considerations: The Study was conducted after taking approval from the Institutional Ethics Committee. Written and informed consent was taken from all study subjects prior to enrolment. The study subjects found to be Vitamin D deficient were referred to hospital specialists for further management.

3. Results

A total of 100 women with T2DM undergoing treatment in the outpatient diabetes clinic at a major tertiary care hospital in Delhi, India were recruited. The mean (±SD) age of the subjects was 47 (±10.8) years. The median (IQR) duration of diabetes since diagnosis was 4 (2.8) years. It was observed that only 22 (22%) of the recruited subjects had good glycemic control (HbA1c < 7%).

The mean (±SD) Vitamin D levels were found to be 15.53 (±10.83) ng/ml. A total of 77 (77%) subjects were Vitamin D deficient of which 40 (40%) were mildly vitamin D deficient, 35 (35%) were moderately deficient and 2 (2%) were severely deficient. Furthermore, there were 16 (16%) subjects with insufficient Vitamin D levels while 7 (7%) were Vitamin D sufficient.

Sociodemographic characteristics: A greater proportion of subjects in the younger age groups (31–45 years) were Vitamin D deficient (86%) compared to older subjects (>60 years) (73.7%) and the difference was statistically significant ($p = 0.02$). Educated women were also significantly less likely to be Vitamin D deficient compared to women who were illiterate. No significant association was found between Vitamin D deficiency and religion or the socioeconomic status of the subjects (Table 1).

Clinical characteristics: The study subjects with BMI between 23 kg/m^2 – 24.9 kg/m^2 (overweight) 22 (95.7%) had vitamin D deficiency while among those who had BMI ≥ 25 kg/m^2, 42 (70%) had Vitamin D deficiency ($p = 0.07$). No significant association was found between Vitamin D deficiency and duration of DM since diagnosis and neither with the inclusion of insulin in treatment (Table 2).

Sunlight exposure: Among those study subjects who had less than one hour of outdoor activity during daytime ($n = 81$), 61 (75.3%) had Vitamin D deficiency, 18.5% of them had Vitamin D insufficiency and 6.2% of them had no deficiency. The difference was not statistically significant compared to the subjects who reported more than an hour of outdoor activity. Similarly, other related factors like duration of direct sun exposure for more than or equal to 45 min during winter, going out in the sun fully covered and application of sunscreens were also not significantly associated with Vitamin D status of the study subjects (Table 2).

Dietary factors: Among the vegetarian subjects ($n = 26$), a total of 17 (65.4%) had Vitamin D deficiency, 8 (30.8) had Vitamin D insufficiency and 1 (3.8) were non deficient in Vitamin D while among the non-vegetarians ($n = 74$), a total of 60 (81.1%) had Vitamin D deficiency, 8 (10.8%) had Vitamin D insufficiency and 6 (8.1%) were non deficient in Vitamin D. The prevalence of Vitamin D deficiency was higher among the subjects who did not consume any milk or fish compared to those who did consume milk or fish but these differences were not statistically significant (Table 3).

Biochemical Parameters: The proportion of subjects with Vitamin D deficiency was higher (79.2%) in those with total cholesterol < 200 mg/dL compared to those with greater cholesterol levels (69.6%) and the difference was statistically significant ($p = 0.01$). However, no significant difference in proportions was found with vitamin D status and LDL levels, HDL levels, TAG levels

Table 1
Distribution of Vitamin D status with their socio-demographic and clinical characteristics among women with type II diabetes mellitus (N = 100).

Characteristics	Vitamin D Non Deficient No(%)	Vitamin D Insufficient	Vitamin D Deficient	Total	P value
Age (In years)					
18–30	0 (0)	3 (60)	2 (40)	5 (100)	0.02*
31–45	3 (6)	4 (8)	43 (86)	50 (100) (100)	
46–59	4 (15.4)	4 (15.4)	18 (69.2)	26 (100)	
≥60	0 (0)	5 (26.3)	14 (73.7)	19 (100)	
Religion					
Hindu	3 (8.3)	7 (19.4)	26 (72.2)	36 (100)	0.69
Muslims	4 (6.3)	9 (14.1)	51 (79.7)	64 (100)	
Education					
Illiterate	6 (7.1)	10 (11.9)	68 (81)	84 (100)	0.03*
Literate	1 (6.3)	6 (37.5)	9 (56.3)	16 (100)	
Socioeconomic status					
Upper Middle	1 (5.0)	3 (15.0)	16 (80)	20 (100)	0.415
Middle	4 (16)	4 (16)	17 (68)	25 (100)	
Lower	2 (3.6)	9 (16.4)	44 (80)	55 (100)	
Occupation					
Homemaker	7 (7.2)	16 (16.5)	24 (76.3)	97 (100)	1
Employed	0 (0)	0 (0)	3 (100)	3 (100)	

Table 2
Association of Vitamin D status and clinical characteristics in women with type II diabetes (N = 100).

Clinical characteristics	Vitamin D Non Deficient n (%)	Vitamin D Insufficient n (%)	Vitamin D Deficient n (%)	Total	P value
DM duration					
≤5 years	2 (3.3)	9 (14.5)	50 (82)	61 (100) (100)	0.17
>5 years	5 (12.8)	7 (17.5)	27 (69.2)	39 (100)	
Type of anti-diabetes medication					
OHA only	5 (6)	14 (16.9)	64 (77.1)	83 (100)	0.64
Insulin	2 (11.8)	2 (11.8)	13 (76.5)	17 (100)	
BMI					
<22.9 kg/m^2	0 (0)	4 (23.5)	13 (76.5)	17 (100)	0.07
23–24.9 kg/m^2	0 (0)	1 (4.3)	22 (95.7)	23 (100)	
≥25 kg/m^2	7 (11.7)	11 (18.3)	42 (70.0)	60 (100)	
Duration of outdoor activity					
<1 Hour	5 (6.2)	15 (18.5)	61 (75.3)	81 (100)	0.29
≥1 Hour	2 (10.5)	1 (5.3)	16 (84.2)	19 (100)	
Duration of direct sun exposure					
<45 min	6 (6.3)	15 (15.6)	75 (78.1)	96 (100)	0.14
≥45 min	1 (25)	1 (25)	2 (50)	4 (100)	
Fully covered in outdoors during daytime					
Yes	6 (9.8)	10 (16.4)	45 (73.8)	61 (100)	0.43
No	1 (2.6)	6 (15.4)	32 (82.1)	39 (100)	
Application of sunscreen lotions					
Yes	0 (0)	0 (0)	2 (100)	2 (100)	1.00
No	7 (7.1)	16 (16.3)	75 (76.5)	98 (100)	

Table 3
Association of dietary risk factors and vitamin D status among the study subjects with type II diabetes mellitus (N = 100).

Characteristics	Vitamin D Non Deficient n (%)	Vitamin D Insufficient n (%)	Vitamin D Deficient n (%)	Total	P value
Diet					
Vegetarian	1 (3.8)	8 (30.8)	17 (65.4)	26 (100)	0.05
Non- vegetarian	6 (8.1)	8 (10.8)	60 (81.1)	74 (100)	
Consumption of Milk					
Yes	4 (7.7)	12 (23.1)	36 (69.2)	52 (100)	0.12
No	3 (6.3)	4 (8.3)	41 (85.4)	48 (100)	
Consumption of yoghurt	5 (8.6)	9 (15.5)	44 (75.9)	58 (100)	0.87
	2 (4.8)	7 (16.6)	33 (78.6)	42 (100)	
Consumption of butter	1 (9.1)	2 (18.2)	8 (72.7)	11 (100)	0.74
	6 (6.7)	14 (15.7)	69 (77.5)	89 (100)	
Consumption of fish	1 (6.2)	5 (31.2)	10 (62.5)	16 (100)	0.19
	6 (7.1)	11 (13.1)	67 (79.8)	84 (100)	
Consumption of egg	5 (9.4)	9 (17.0)	39 (73.6)	53 (100)	0.59
	2 (4.2)	7 (14.9)	38 (80.9)	47 (100)	

and fasting blood glucose levels. No relationship was also found between Vitamin D status and glycemic control (Table 4). The mean (±SD) levels of HbA1c among the subjects having Vitamin D deficiency, insufficiency and sufficiency was 8.87 (±1.87), 9.25 (±2.51) and 9.82 (±1.36) respectively. The difference was not found to be statistically significant. There also existed no correlation ($r = 0.003$) between the Vitamin D levels and HbA1c levels.

4. Discussion

The present study observed that more than three fourths of women with T2DM had vitamin D deficiency (77%) and poor glycemic control (78%). The Vitamin D deficiency estimates in our study are similar to that in another hospital based study conducted by Hari Narayan et al. in South India (2011) among 136 women where vitamin D deficiency prevalence was estimated as 76% in women of the reproductive age group and 70% in the post-menopausal group [18]. However, the vitamin D deficiency prevalence found in our study is lower compared to some other studies. A hospital study by Sheth et al. (2014) [19] from Gujarat in Western India found 91.4% vitamin D deficiency in women diabetes subjects while another study by Zahrani et al. (2013) in Saudi Arabia reported most (98.4%) diabetes subjects being Vitamin D deficient [20].

No relationship was found between Vitamin D levels and glycemic control status. This finding is in agreement with previous Indian studies [19,21]. So we infer that vitamin D deficiency is not a determinant of glycemic control in Indian women with T2DM. A pilot randomized control trial (RCT) in Mumbai, India (2010) among 28 Type 2 DM patients found that short-term improvement in vitamin D status after 4 months of daily supplementation did not improve HbA1c levels [22]. There exists insufficient evidence that Vitamin D deficiency is related with glycemic status in women diabetes patients. Instead, in resource-constrained settings it is more likely that factors like patient medication adherence status and the extent of clinical inertia determine glycemic control status [23].

Our study found that Vitamin D deficiency was more likely in younger women with diabetes compared to older women. Moreover, women with lower self-reported sun-exposure had a higher prevalence of Vitamin D deficiency. This finding is in agreement with the results of a previous study from the Punjab state in North India by Singh et al. (2010) [24]. Lack of sun exposure in younger women due to conservative cultural traditions which promote full-bodied clothing and also concerns regarding darkening of skin complexion can explain such a finding.

In the present study, vitamin D deficiency was more prevalent in non-vegetarian subjects compared to vegetarian women. Non-vegetarian diet is expected to protect against Vitamin D deficiency. However, our study findings could be due to the demographic profile of our study population, which consisted predominantly of Muslim women who while mostly non-vegetarian but also had lower sun-exposure due to wearing of traditional full-bodied clothing.

It was observed in the present study that subjects with high TAG levels had more Vitamin D deficiency. However, an earlier study from Korea observed positive correlation between vit D level and serum lipid profile [25]. This difference may be because of differing baseline population characteristics and the genetic distinction. Furthermore, our study found that subjects with hypercholesterolemia were significantly less likely to be Vitamin D deficient. It is known that blood-cholesterol reaches the skin and is converted into Vitamin D on exposure to UV rays present in sunlight. Hence, the lack of cholesterol could ultimately lead to a Vitamin D deficient state. This indicates the need for judicious application of statins in diabetes patients [26].

We conclude that vitamin D deficiency is highly prevalent among women with T2DM. Younger age, illiteracy, vegetarianism and low serum cholesterol level might be the major determinants of vitamin D deficiency. However, except for blood cholesterol, vitamin D deficiency probably does not influence other biochemical parameters linked with diabetes health outcomes including the glycemic control.

5. Limitations

There exist certain limitations in the study. The small sample size of the study precludes the application of regression analysis to detect confounding variables. Furthermore, the cross-sectional study design rules out the possibility of finding out any temporal association between Vitamin D levels and biochemical parameters linked to health outcomes in diabetes patients. Since the study was conducted only among women with T2DM, the results cannot be generalized to the male gender. Future studies with larger study samples and with a prospective longitudinal study design could provide more conclusive evidence.

Table 4
Vitamin D status association with various Biochemical Parameters (N = 100).

Biochemical parameters	Vitamin D Non Deficient n (%)	Vitamin D Insufficient n (%)	Vitamin D Deficient n (%)	Total	P value
Fasting plasma Glucose					
<126 mg/dL	1 (3.3)	6 (20)	23 (76.7)	30	0.53
≥126 mg/dL	6 (8.6)	10 (14.3)	54 (77.1)	70	
Glycated haemoglobin (HbA1c)					
HbA1c < 7%	0 (0)	4 (18.2)	18 (81.8)	22	0.46
HbA1c ≥ 7%	7 (9)	12 (15.4)	59 (75.6)	78	
LDL Cholesterol					
<100 mg/dL	1 (2)	6 (12)	43 (86)	50	0.06
≥100 mg/dL	6 (12)	10 (20)	34 (86)	50	
HDL Cholesterol					
<50 mg/dL	5 (6.4)	12 (15.6)	60 (78)	77	0.76
≥50 mg/dL	2 (8.6)	4 (17.4)	17 (74)	23	
Triglycerides (TAG)					
<50 mg/dL	2 (3.8)	10 (19.2)	40 (77)	52	0.37
≥150 mg/dL	5 (10.4)	6 (12.6)	37 (77)	48	
Total Cholesterol					
<200 mg/dL	2 (2.6)	14 (18.2)	61 (79.2)	77	0.01*
≥200 mg/dL	5 (2.2)	2 (8.7)	16 (69.7)	23	

Sources of support

Nil.

Conflicts of interest

None to declare.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.dsx.2019.03.005.

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