ASSOCIATION OF VITAMIN D LEVEL WITH MICRO-VASCULAR COMPLICATIONS OF TYPE 2 DIABETES MELLITUS

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ABSTRACT

Introduction: In diabetes, role of vitamin D in insulin sensitivity and its protective effect from complications has been proven by many animal model studies. However, place of vitamin D as a preventive or therapeutic measure in diabetes mellitus is yet not established. **Aim:** To study the association between vitamin D level and micro vascular complications of diabetes mellitus. **Methods:** A hospital based observational study on 119 Patients of type 2 diabetes mellitus at a tertiary care center India was done between Jan 2020 to Dec 2022. All routine lab parameters were done and patients were evaluated for diabetic retinopathy and neuropathy and nephropathy. Statistical analysis was done by pearson correlation test, fisher's exact test and ANOVA. Linear regression analysis was also done. **Results:** out of 119 patients, with mean age of 49.5 year, mean duration of diabetes 6.5yr. On comparison between vitamin D level and frequency of diabetic neuropathy, significant association was found whereas no significant association was found between vitamin D deficiency and diabetic nephropathy and retinopathy. **Conclusion:** Vitamin D deficiency may be a risk factor for development of diabetic neuropathy and dyslipidaemia, in patients with type 2 diabetes mellitus.

Keywords: Vitamin D, Diabetes mellitus, complications

INTRODUCTION

In recent years, role of vitamin D in beta cell function, insulin secretion and insulin resistance has been studied. In 1980s, some studies were done on rat pancreas to assess role of vitamin D in insulin secretion. Insulin secretion was compared between vitamin D deficient and vitamin D fed rats, concluding that vitamin D supplementation seemed to improve insulin secretion.[1-3]

The role of vitamin D, not only is described in diabetes mellitus, but also in its various micro vascular and macro vascular complications. In diabetic nephropathy various animal model studies suggest that vitamin D can prevent proteinuria and podocyte injury.[4,5] Diabetic retinopathy is also a common micro vascular complication of diabetes, yet its treatment and preventive measures are insufficient. The pathophysiologic mechanisms involved in diabetic retinopathy are loss of retinal pericytes, increased retinal vascular permeability, alterations in retinal blood flow and abnormal retinal microvasculature.[6] Vitamin D receptor is present on endothelial cells, pericytes and vascular smooth muscle cells and it exerts vasculo-protective function.[33]Role of vitamin D is also described in another micro vascular complication of diabetes mellitus i.e. diabetic neuropathy. Various neurodegenerative disorders such as Parkinson's disease, multiple sclerosis and cognitive dysfunction has been linked with vitamin D deficiency in past studies.[7]

In diabetes, its role in insulin sensitivity and its protective effect from complications has been proven by many animal model studies. However, place of vitamin D as a preventive or therapeutic measure in diabetes mellitus is yet not established. Vitamin D deficiency, being the prevalent condition in Indian population, is needed to be explored in the association with other metabolic conditions such as diabetes mellitus.

Aim: To study the association between vitamin D level and micro vascular complications of diabetes mellitus.

Methods: A hospital based observational study on 119 Patients of type 2 diabetes mellitus at **Department of Medicine at Government institute of medical sciences, Gautam Buddha nagar (UP),** India was done between Jan 2020 to Dec 2022. All routine lab parameters were done and patients were evaluated for diabetic retinopathy and neuropathy and nephropathy. Statistical analysis was done by pearson correlation test, fisher's exact test and ANOVA. Linear regression analysis was also done. All cases of type 2 diabetes mellitus of all age group attending opd or ipd will be included. Patients with established chronic kidney disease, chronic liver disease, carcinoma, on glucocorticoid therapy, diuretics, anti-epileptic drugs or supplementations of vitamin D, having acute complications like diabetic ketoacidosis or hyperglycaemic hyperosmolar state, severe anaemia or needed blood transfusions were excluded from study.

Data collection

After taking approval from scientific research committee (SRC) and institutional ethics committee (IEC) and fulfilling inclusion and exclusion criteria patients were selected. All sociodemographic information and routine lab parameters were done. Kidney function test (serum urea, serum creatinine, and serum uric acid), urine albumin to creatinine ratio (UACR), fasting lipid profile, fasting insulin and 25-hydroxy vitamin D level were also done. Patients were assessed for diabetic

neuropathy by clinical examination. Patients were assessed for diabetic retinopathy by fundus examination.

Statistical analysis

Thus collected data was entered in Microsoft excel sheet and analysed by Epi info software by CDC

RESULTS

In our study out of 119 cases maximum 67 were male, 70 cases were urban and mean age of population was 49.5 ± 11 year. Mean duration of diabetes was 6.5 ± 5.18 year. Out of these, 22 patients had history of smoking and 11 patients had history of alcohol intake. On the education background, 26 patients were illiterate, 65 patients received primary education while 28 patients had graduation level of education. 30 patients were found to have family history of diabetes mellitus in first degree relatives.

Significant association was found between age and vitamin D. (fisher's exact = 0.012) indicating vitamin D deficiency is more common in young population. Nearly significant association was found between sex and residence and vitamin D level. (Fisher's exact = 0.084 and 0.081, respectively)

Table1. Sociodemographic profile and correlation with Vit D

Sociodemographic variable		**Vitamin D sufficient (>20 ng/ml)**	**Vitamin D deficient (<=20 ng/ml)**	**Fisher's exact**
Age (n=119)	<=40 year	4	24	0.012
	>40 year	38	53	
Sex (n=119)	Male	19	48	0.084
	Female	23	29	
Residence (n=119)	Urban	20	50	0.081
	Rural	22	27	
Smoking (n=119)	Yes	7	15	0.808
	No	35	62	
Alcohol (n=119)	Yes	5	6	0.516
	No	37	71	
Education (n=119)	Illiterate	11	15	0.145
	Primary	18	47	
	Graduation	13	15	
BMI (n=119)	Underweight	1	0	0.305
	Normal	7	7	
	Overweight	6	10	
	Obese	28	60	

Correlation of serum 25 (OH) D levels was done with selected variables. A significant positive correlation was seen between vitamin D level and age (r = 0.1824, p = 0.0471). HbA1c and fasting

plasma glucose were found to be negatively correlated with vitamin D level (r = -0.0994, p = 0.2824) and (r = -0.1401, p = 0.1285), respectively, however, not significant. There was negative but non-significant relation found between vitamin D level and total cholesterol (r = -0.0134, p = 0.8987), serum triglyceride level (r = -0.0310, p = 0.7690) and serum LDL (r = -0.0149, p = 0.8873). Vitamin D had negative correlation with serum creatinine (r = -0.0944, p = 0.3707), serum uric acid (r = -0.0834, p = 0.4295), UACR (r = -0.0188, p = 0.8709) but on the contrary, also had negative correlation with eGFR (r = -0.0073, p = 0.9447), however, correlations were not significant. A significant positive correlation was present between vitamin D level and serum albumin (r = 0.2607, p = 0.0195).

Table2. Biochemical profile and Correlation of vitamin D level with different parameters

S.no.	Parameter	Frequency (n)	Mean	Standard deviation	Correlation with Serum 25 (OH) D r(p)
1.	Age	119	49.4958	11.00452	0.1824 (0.0471)
2.	BMI	119	27.1042	4.21437	0.0367 (0.6919)
3.	HbA1c	119	9.061345	2.308279	-0.0994 (0.2824)
4.	Fasting plasma glucose	119	183.0277	39.17798	-0.1401 (0.1285)
6.	Serum 25 (OH) vitamin D	119	20.10706	16.2246	-
7.	Fasting insulin	111	10.5018	9.08206	0.0817 (0.3938)
8.	Total cholesterol	93	171.9108	39.63992	-0.0134 (0.8987)
9.	Serum triglyceride	92	166.6728	100.4567	-0.0310 (0.7690)
10.	Serum HDL	93	56.66129	14.39644	0.0296 (0.7782)
11.	Serum LDL	93	95.92065	36.66769	-0.0149 (0.8873)
13.	Serum creatinine	92	1.001087	0.2995398	-0.0944 (0.3707)
14.	Serum uric acid	92	5.342391	1.810666	-0.0834 (0.4295)
15.	eGFR	92	85.0762	22.97076	-0.0073 (0.9447)
16.	UACR	77	601.5536	1201.938	-0.0188 (0.8709)
17.	Serum albumin	80	4.368875	0.3406843	0.2607 (0.0195)
21.	Serum TSH	82	2.392195	1.763995	0.0456 (0.6839)
22.	CRP	59	4.789322	5.587212	

For comparison, patients were divided into different subgroups on the basis of different variables. Patients with serum 25(0H) D <=20 ng/ml were considered as vitamin D deficient (64.71%) while 35.29 % cases had serum 25 (OH) D > 20 ng/ml were considered as vitamin D sufficient.

112 patients were evaluated for diabetic nephropathy. Out of these, 68 patients (60.71%) were found to have diabetic nephropathy. When compared with vitamin D status, out of 68 patients with diabetic nephropathy, 43 patients (63.23%) were found to have vitamin D deficiency, however association was not significant. (Fisher's exact = 0.845)

85 patients were examined for diabetic neuropathy. Out of these, 37 patients (43.53%) were found to have diabetic neuropathy. When compared with vitamin D status, out of 37 patients with diabetic neuropathy, 27 patients (73%) were found to have vitamin D deficiency. Significant association was found between vitamin D deficiency and diabetic neuropathy. (Fisher's exact = 0.045)

74 patients were examined for diabetic retinopathy. Out of these, 32 patients (43.24%) were found to have diabetic retinopathy. When compared with vitamin D status, out of 32 patients with diabetic retinopathy, 22 patients (68.75%) were found to have vitamin D deficiency, however association was not significant. (Fisher's exact = 0.342)

Table3. Vitamin D status with diabetic micro vascular complications

Microvascular complications		**Vitamin D sufficient (>20 ng/ml)**	**Vitamin D deficient (<=20 ng/ml)**	**Fisher's exact**
Diabetic nephropathy (n=112)	Yes	25	43	0.845
	No	17	27	
Diabetic neuropathy (n=85)	Yes	10	27	0.045
	No	24	24	
Diabetic retinopathy (n=74)	Yes	10	22	0.342
	No	18	24	

Mean serum triglyceride level in vitamin D deficient, insufficient and sufficient groups were 206 ± 147.3 mg/dl, 152.7 ± 64 mg/dl and 147.8 ± 72.6 mg/dl respectively. Vitamin D deficiency was found to be associated with elevated serum triglyceride level, but association was near significant. (P-value = 0.0510)

Table4. Lipid profile and Vit D correlation (ANOVA test)

S.no.	**Lipid Profile**	**Vitamin D deficient (<=10 ng/ml) (n=38)**	**Vitamin D insufficient (11-20 ng/ml) (n=39)**	**Vitamin D sufficient (>20 ng/ml) (n=42)**	**P-value**
1.	Total cholesterol	181.78571 ± 46.33242 (n=28)	167.09091 ± 37.833817 (n=33)	168.24062 ± 34.4862 (n=32)	0.2895
2.	Serum triglyceride	205.96296 ± 147.30854 (n=27)	152.78788 ± 64.075033 (n=33)	147.84062 ± 72.593876 (n=32)	0.0510
3.	Serum HDL	60.728571 ± 14.955857 (n=28)	52.139394 ± 14.384148 (n=33)	57.765625 ± 12.98944 (n=32)	0.0569
4.	Serum LDL	89.475714 ± 33.960089 (n=28)	101.56061 ± 40.514997 (n=33)	95.74375 ± 34.926845 (n=32)	0.4437

DISCUSSION

In our study, serum creatinine and UACR both showed negative correlation with Vitamin D level, however association was not significant. On contrary, a negative insignificant correlation was found between vitamin D and eGFR. On subgroup analysis, 112 patients were evaluated for diabetic nephropathy. 60.71% patients were found to have diabetic nephropathy. No significant association was found between vitamin D and diabetic nephropathy. Similarly **Xiao Y *et al*[8]** in 2020 conducted a study and found no significant association between vitamin D deficiency and diabetic nephropathy and retinopathy. Also **Girard E *et al*[9]** in 2021 conducted an observational study and no association was found between vitamin D deficiency and diabetic nephropathy.

In our study, a total of 85 patients were evaluated for diabetic neuropathy by clinical examination. Out of these, 37 patients (43.53%) were found to have diabetic neuropathy. When frequency of diabetic neuropathy was compared between vitamin D sufficient and Vitamin D deficient groups, vitamin D deficiency was significantly associated with diabetic neuropathy. Similarly **Abdelsadek SE *et al*[10]** in 2018 conducted a study to compared vitamin D level between patients with diabetic neuropathy and without diabetic neuropathy witch was found to be significant. **He R *et al*[11]**, **Fan L *et al*[12]** also supported the association of vitamin D deficiency with diabetic neuropathy. **Zhang B *et al*[13]** in 2019 conducted a systematic review and meta-analysis to assess the relation of vitamin D deficiency with diabetic neuropathy. They included 75 studies for systematic review and 13 studies were selected for meta-analysis. Consistent with our result, significant association was found between vitamin D deficiency and diabetic neuropathy.

Previously, vitamin D deficiency has been linked to various neurodegenerative disorder such as Parkinson's disease, multiple sclerosis and cognitive dysfunction.[7] Possible mechanisms by which vitamin D has a role to play in delaying of neurodegeneration are up regulation of neurotrophic factors such as nerve growth factor (NGF) and glial cell line derived neurotrophic factor (GDNF) and maintenance of neuronal calcium homeostasis.[14-16]

In our study, a total of 74 patients were evaluated for diabetic retinopathy, out of these, 32 patients (43.24%) were found to have diabetic retinopathy. When compared with vitamin D status, no significant association was found between vitamin D deficiency and diabetic retinopathy. In consistency with our result **Alam U *et al*[17]** in their study, no significant association was found between vitamin D level and diabetic retinopathy.

In addition, in our results, a negative correlation was found between vitamin D level and total cholesterol (r = -0.0134), serum triglycerides (r = -0.0310) and serum LDL (r = -0.0149) while a positive correlation was found between vitamin D level and serum HDL (r = 0.0296). When mean values of lipid profile parameters were compared among vitamin D deficient (<=10 ng/ml), vitamin D insufficient (11-20 ng/ml) and vitamin D sufficient (>20 ng/ml), near significant association was found between vitamin D deficiency and hypertriglyceridemia. (p = 0.0510) Similarly **Bando H *et al*[18]** in their study they found significant association between vitamin D deficiency and increased triglyceride level. Also **Saedisomeolia A *et al*[19]** conducted a cross sectional study to assess the relation of vitamin D level and lipid profile in type 2 diabetes mellitus. In consistence with our result,

they found negative correlation of vitamin D with total cholesterol, serum triglyceride and serum LDL level and positive correlation was found between vitamin D level and serum HDL level. Significant association was found between vitamin D deficiency and raised serum triglyceride level.

CONCLUSION

Vitamin D deficiency may be a risk factor for development of diabetic neuropathy in patients with type 2 diabetes mellitus. Vitamin D deficiency may be related to dyslipidaemia in type 2 diabetes mellitus. Vitamin D deficiency is associated with hypertriglyceridemia.

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