How to Cite:
Najmi, M. A., Zaka-ur-Rab, S., Arrin, S., & Ali, A. (2022). Relationship between serum vitamin D levels in diabetic patients with and without diabetic retinopathy. *International Journal of Health Sciences*, *6*(S8), 3026–3033.
https://doi.org/10.53730/ijhs.v6nS8.12744

Relationship between serum vitamin D levels in diabetic patients with and without diabetic retinopathy

Dr Mohammad Abubakar Najmi
MS, Ex Resident, Institute of Ophthalmology, Jawaharlal Nehru Medical College, Aligarh Muslim University, Aligarh, 202002, Uttar Pradesh, India
Email: abubakar.najmi@gmail.com

Dr Simi Zaka-ur-Rab
MS, DNB, MNAMS, Professor, Aligarh Muslim University Institute of Ophthalmology, Jawaharlal Nehru Medical College, Aligarh, 202002, Uttar Pradesh, India
Corresponding author email: s_zrab@yahoo.co.in

Dr Shazia Arrin
MS, Assistant Professor, Aligarh Muslim University Institute of Ophthalmology, Jawaharlal Nehru Medical College, Aligarh, 202002, Uttar Pradesh, India
Email: shazia.ar2114@gmail.com

Dr Asif Ali
Phd, Professor, Department Of Biochemistry, Jawaharlal Nehru Medical College, Aligarh Muslim University, Aligarh-202002, Uttar Pradesh, India
Email: asifali_amu@rediffmail.com

***Abstract*---**Aim: To study the relationship between serum Vitamin D levels and severity of Diabetic Retinopathy. Materials and Methods: This cross-sectional study was conducted on 346 subjects. The cases were divided into three groups, group A comprised of 180 type 2 diabetic patients with retinopathy, group B comprised of 86 type 2 diabetic patients without retinopathy (no DR) and group C comprised of age and gender matched 80 healthy non diabetic controls. Group A further divided into group A1 consisting 84 proliferative diabetic retinopathy (PDR) patients and group A2 consisting of 96 non proliferative diabetic retinopathy patients (NPDR). Results: The difference in the levels of serum Vitamin D between group A1 and group B and group A1 and group C were statistically significant with p value and Confidence Interval (CI) being (<0.001, -10.35 to -4.66) and (<0.001, -11.45 to -5.54) respectively. There was statistically significant negative correlation between HbA1c and serum Vitamin D

in diabetic patients with PDR (r=-0.464, p=0.002), diabetic patients with NPDR (r=-0.399, p=0.005) and diabetic patients with no DR(r=-0.397, p=0.011). Conclusions: Serum vitamin D level decreases significantly with increasing severity of Diabetic Retinopathy and increasing levels of HbA1c.

***Keywords*---**vitamin D, diabetes, diabetic retinopathy, glycaemic control.

Introduction

Diabetes mellitus is a group of metabolic diseases characterised by hyperglycaemia resulting from defect in insulin secretion, insulin action or both. Diabetic retinopathy (DR) is a severe complication of diabetes and the leading cause of blindness among working adults worldwide. In 2010, of an estimated 285 million people worldwide with diabetes, over one-third have signs of DR, and a third of these are afflicted with vision-threatening diabetic retinopathy (VTDR), defined as severe Non Proliferative Diabetic Retinopathy (NPDR) or Proliferative Diabetic Retinopathy (PDR) or the presence of diabetic macular oedema. [1] India is fast becoming the hub of diabetes so the number of patients with the complications of diabetes like Diabetic Retinopathy with increase in near future.
Vitamin D has important actions on glucose metabolism. These include improved insulin exocytosis, direct stimulation of insulin receptor, improved uptake of glucose by peripheral tissues, improving insulin resistance.[2],[3],[4] There is a growing interest on the potential role of vitamin D in the development of diabetic micro and macroangiopathic complications.[5],[6] The link between inflammation and diabetic morbidities on one hand and between low circulating 25-hydroxy Vitamin D[25(OH)D] and inflammation on the other hand have been recently reported.[7],[8]

A potential association between vitamin D and the presence of this microangiopathic complication in patients with type 2 diabetes has been reported by various authors.[9],[10],[11],[12],[13],[14] Alcubierre *et al* (2015), [15]Ashinne *et al (*2018)[16] and Nadri et al (2021) [17] have reported the association of vitamin D deficiency with the presence and severity of diabetic retinopathy in type 2 diabetes. Zoppini*et al*,[18] in 2015, reported an inverse and independent relationship between circulating 25(OH)D levels and the prevalence of microvascular complications in patients with type 2 diabetes. In 2019 Nadri *et al*, reported that serum vitamin D levels of ≤ 18.6 ng/mL serve as sensitive and specific indicator for proliferative disease, among patients of diabetic retinopathy.[19] In a study conducted by Trott *et al* in 2022 , authors found that Vitamin D deficiency is significantly associated with sight threatening diabetic retinopathy (including DME), but not with non sight threatening diabetic retinopathy.[20] Given the well-reported associations between low serum vitamin D levels and other unfavourable outcomes, it is important that low serum vitamin D levels is managed appropriately and in a timely manner to reduce the risk of blindness in diabetic patients.

Materials and Methods

Study Population

The present study was undertaken on 346 patients. All the Tenets of the Declaration of Helsinki were adhered to. A well informed written consent for blood sampling was taken from all the patients included in the study. Patients with systemic malignancy, infection, inflammatory disorder and patients who have had, ocular trauma, infective ocular disease, received intravitreal anti-VEGF injection, Corticosteroids within past 4 weeks, Vitamin / mineral supplements within past 4 weeks, and eyes with significant corneal opacities, dense cataract, and significant vitreous haze were excluded. Patients on Vitamin D supplementation were also excluded.

The cases were divided into 3 groups:

- Group A: This group served as the study group and comprised of 180 type 2 diabetic patients with retinopathy. This was further divided into group A1 consisting 84 proliferative diabetic retinopathy patients and group A2 consisting of 96 non proliferative diabetic retinopathy patients. In diabetic patients having different grades of diabetic retinopathy in either eye, then the eye with severe grade of diabetic retinopathy was considered for defining the case.
- Group B: This group served as study group and comprised of 86 diabetic patients without retinopathy.
- Group C: This group served as the control group and comprised of 80 age and gender matched healthy patients who were not suffering from diabetes.

Data Collection

All the patients were evaluated clinically and details were documented. A thorough clinical history was taken from all the patients. The patients were thoroughly examined. Best corrected visual acuity, intraocular pressure was recorded at baseline. Slit lamp examination of the anterior segment was done. Fundus was evaluated in detail with a +90D lens and Goldman three mirror contact lens. A detailed direct and indirect ophthalmoscopy was performed. Fluorescein angiography was performed wherever required. Diabetic Retinopathy was identified according to the lesion defined by the Early Treatment Diabetic Retinopathy Study severity scale was present; these included microaneurysm, hemorrhage, hard exudate, cotton wool spots, intraretinal microvascular abnormalities, venous beading, and new vessels.

All the blood samples for the analysis were properly collected by venipuncture technique without application of any tourniquet (to avoid even minimal chances of hemolysis) from cubital fossa of either arm after cleansing the site with spirit swab. The concentration of Vitamin D was measured in the serum using Beckman Coulter Access 2 System, Beckman Coulter, Inc USA. This system is a paramagnetic particle, chemiluminescent immunoassay for the quantitative determination of total 25-hydroxyvitamin D [25(OH) vitamin D] levels in human serum.

Statistical Analysis

Statistical analysis was done using SPSS 21.0 for Windows software (SPSS Inc., Chicago, Illinois, USA). Independent samples t-tests were applied for analysing statistical significance of changes in levels of HbA1c and serum vitamin D in between the groups. Pearson correlation coefficient was used to find out any correlation between HbA1c level and serum vitamin D level within the groups. P value <0.05 was taken as significant.

Results

The age of patients ranged from 34 years to 77 years with mean age of 52.66±10.90 years. Out of 346 cases enrolled in the study 162 (46.8%) were females and 184 (53.2%) were males. The age of the patients ranged in Group A1 from 27 to 66 years with the mean age of 50.32±9.71 years. Out of 84 patients enrolled in group A1, 55(65.5%) were males and 29(34.5%) were females. The age of the patients ranged in Group A2 from 34 to 76 years with the mean age of 53.71±11.96 years. Out of 96 patients enrolled in group A2, 47(49.0%) were males and 49(51.0%) were females. The age of the patients in group B ranged from 35 to 77 years with the mean age of 53.27±11.06 years. Out of 86 patients enrolled in Group B, 50 (58.1%) were males and 36 (41.9%) were females. The age of the patients in group C ranged from 35 to 75 years with the mean age of 53.00±11.13 years. Out of 80 patients enrolled in Group C, 32 (40.0%) were males and 48(60.0%) were females (Table 1).

Table 1: Age and Gender Wise Distribution of Patients

GROUP	NUMBER OF PATIENTS	GENDER		AGE (Years)	
		MALE	FEMALE	RANGE	MEAN±SD
A1 (PDR)	84	55	29	27-66	50.32±9.71
A2 (NPDR)	96	47	49	34-76	53.71±11.96
B (NO DR)	86	50	36	35-77	53.27±11.06
C (CONTROL)	80	32	48	35-75	53.00±11.13
TOTAL	346	184	162	27-66	50.32±9.71

Mean levels of Vitamin D, HbA1c and Serum Calcium among the various groups are depicted in Table 2.

Table 2: Serum Vitamin D, HbA1c and Serum Calcium Levels in Various Groups

Groups	Mean±SD		
	Serum Vitamin D (ng/mL)	HbA1c (%)	Serum Calcium (mg/dL)
Group A1 (Proliferative Diabetic	16.26±6.10	8.55±1.32	8.30±0.52

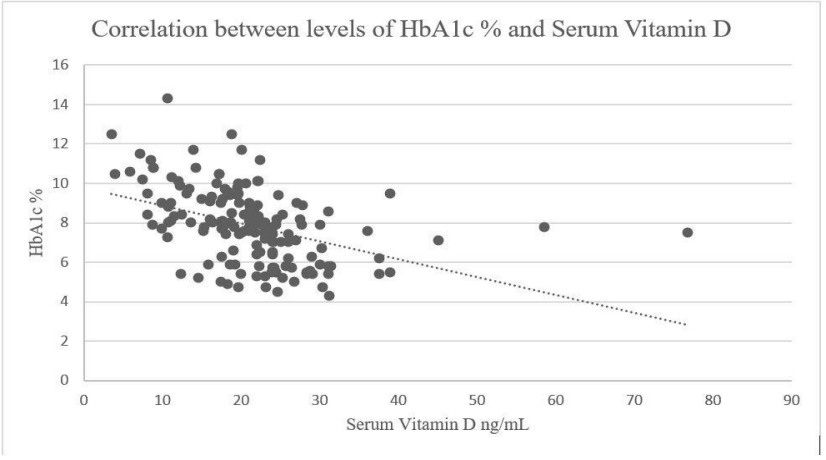
Retinopathy)			
Group A2 (Non Proliferative Diabetic Retinopathy)	20.83±11.67	8.92±1.79	8.65±0.68
Group B (No Diabetic Retinopathy)	23.77±6.73	7.72±1.49	8.73±0.51
Group C (Controls)	24.76±6.25	5.50±0.59	8.70±.83

Mean serum Vitamin D levels decrease with increasing severity of Diabetic Retinopathy whereas mean HbA1c levels increases with the increasing severity of Diabetic retinopathy. The difference in the levels of serum Vitamin D between group A1 and group A2, group A1 and group B and group A1 and group C were statistically significant with p value and Confidence Interval (CI) being (0.02, -8.59 to -0.55), (<0.001, -10.35 to -4.67) and (<0.001, -11.45 to -5.54) respectively. Furthermore the difference in the levels of HbA1c between group A1 and group B, group A1 and group C, group A2 and group B, group A2 and group C and group B and group C were statistically significant with p value and CI being (0.007, 0.23 to 1.46), (<0.001, 2.62 to 3.52), (0.001, 0.49 to 1.89), (<0.001, 2.86 to 3.97) and (<0.001, 1.70 to 2.74) respectively. There is statistically significant negative correlation between HbA1c and Serum Vitamin D levels in groups A1, A2 and B (Table 3 and Fig 1).

Table 3: Correlation between HbA1c and Serum Vitamin D in Various Groups

Groups	R	p
Group A1 (PDR)	-0.464	0.002
Group A2 (NPDR)	-0.399	0.005
Group B (No DR)	-0.397	0.011
Group C (Controls)	-0.115	0.544

Fig 1: Correlation between levels of HbA1c% and Serum Vitamin D



Discussion

This study demonstrates that the serum Vitamin D levels decrease with poor glycaemic control denoted by high HbA1c levels and increasing severity of Diabetic Retinopathy. Furthermore, serum Vitamin D and HbA1c levels showed statistically significant negative correlation. Consistent with our study Akshoy *et al*,[9]in 2000 reported that the mean 1,25(OH)2D concentrations fell with increasing severity of DR, being 63.4 ± 17.26 pmol/L for background DR, 47.7 ± 13.27 pmol/L for preproliferative DR, and 43.1 ± 19.45 pmol/L for proliferative DR. Compared with the control group, serum 25(OH)D concentrations were found to be decreased in diabetic patients ($p < 0.001$). Ashinne *et al* in 2018 reported that lower serum 25(OH)D was associated with increased severity of DR and the presence of Vitamin D deficiency was associated with a two-fold increased risk for proliferative DR.[16]

Ahmadieh *et al*, in 2013 reported that the mean 25-OHD levels were lower in subjects with diabetic retinopathy compared to those without retinopathy (12.3±5.5 versus 21.8±13.7, $p <0.001$).[10] And Alcubierre *et al*,[15]in 2015, found that 25(OH)D concentrations were lower in diabetic patients with retinopathy compared to diabetic patients without retinopathy, this difference reached only borderline statistical difference ($p= 0.05$). In 2019 Nadri *et al*, reported that serum vitamin D levels of ≤ 18.6 ng/mL serve as sensitive and specific indicator for proliferative disease, among patients of diabetic retinopathy.[18]

We observed statistically significant negative correlation between HbA1c and serum Vitamin D in diabetic patients with PDR ($r=-0.464$, $p=0.002$), diabetic patients with NPDR($r=-0.399$, $p=0.005$) and diabetic patients with no DR($r=-0.397$, $p=0.011$). Ahmadieh *et al*,[10] has also reported negative correlation between serum 25-OHD and HbA1c levels ($r = -0.02$, $p=0.049$). He *et al*, in 2014 reported a downward trend in average 25–hydroxyvitamin D level with the increased stages of diabetic retinopathy ($p < 0.01$).[13] Moreover in 2017 Long *et al*,[21] reported that poorly controlled HbA1c is associated with greater severity of diabetic retinopathy.

Conflicting Interest : Nil
Acknowledgement: None

References

1. Ahmadieh H, Azar ST, Lakkis N, Arabi A. Hypovitaminosis D in patients with type 2 diabetes mellitus: a relation to disease control and complications. *ISRN Endocrinology*. 2013;641098:7.
2. Aksoy H, Akcay F, Kurtul N, Baykal O, Avci B. Serum1,25 dihydroxy vitamin D (1,25(OH)2D3), 25 hydroxy vitamin D(25(OH)D) and parathormone levels in diabetic retinopathy. *Clinical Biochemistry*. 2000; 33(1):47–51.
3. Alcubierre N, Valls J, Rubinat E, Cao G, Esquerda A, Traveset A *et al*. Vitamin D Deficiency Is Associated with the Presence and Severity of Diabetic Retinopathy In Type 2 Diabetes Mellitus. J Diabetes Res 2015;374178:7.

4. Ashinne B, Rajalakshmi R, Anjana RM, Narayan KMV, Jayashri R*et al*. Association of serum vitamin D levels and diabetic retinopathy in Asian Indians with type 2 diabetes. Diabetes Res Clin Pract. 2018;139:308-313.
5. He R, Shen J, Liu F, Zeng H, Li L, Yu H*et al*. Vitamin D deficiency increases therisk of retinopathy in Chinese patients with Type 2 diabetes. *Diabetic Medicine*. 2014; 31(12):1657–1664.
6. Hess K, Grant PJ. Inflammation and thrombosis in Diabetes. Thrombosis and Haemostasis Supplement. 2011; 1:43-50.
7. Jablonski KL, Chonchol M, Pierce G L, Walker AE, Seals DR. 25-HydroxyvitaminD deficiency is associated with inflammation-linked vascular endothelial dysfunction in middle aged and older adults.Hypertension. 2011; 57(1):63-9.
8. Joergensen C, Hovind P, Schmedes A, Parving HH, Rossing P. Vitamin D levels, microvascular complications, and mortality in type 1 diabetes. *Diabetes Care.* 2011; 34(5): 1081–1085.
9. Joergensen C, Reinhard H, Schmedes A, Hansen PR, Wiinberg N, Petersen CL et al. Vitamin D levels and asymptomatic coronary artery disease in type 2 diabetic patients with elevated urinary albumin excretion rate. *Diabetes Care*. 2012; 35(1):168–172.
10. Kadowaki S, Norman AW. Dietary vitamin D is essential for normal insulin secretion from the perfused rat pancreas. J. Clin. Invest. 1984; 73:759–766.
11. Long M, Wang C, Liu D. Glycated hemoglobin A1C and vitamin D and their association with diabetic retinopathy severity. Nutrition & Diabetes. 2017; 7:281.
12. Mitri J, Dawson-Hughes B, Hu FB, Pittas AG. Effects of vitamin D and calcium supplementation on pancreatic β cell function, insulin sensitivity, and glycemia in adults at high risk of diabetes: The Calcium and Vitamin D for Diabetes Mellitus (CaDDM) randomized controlled trial. 2011; 94:486–94.
13. Nadri G, Saxena S, Kaur A, Ahmad K, Garg P *et al*. Correlation between vitamin D serum levels and severity of diabetic retinopathy in patients with type 2 diabetes mellitus. Journal of Endocrinology, Metabolism and Diabetes of South Africa 2021; 26(3):82–88
14. Nadri G, Saxena S, Mahdi AA, Kaur A, Ahmad KM *et al*. Serum vitamin D is a biomolecular biomarker for proliferative diabetic retinopathy. International Journal of Retina and Vitreous. 2019; 5:31.
15. Need AG, O'Loughlin PD, Horowitz BE, Nordin C. Relationship between fasting serum glucose, age, body mass index and serum 25 hydroxy vitamin D in postmenopausal women. Clinical Endocrinology. 2005; 62(6):738-41.
16. Patrick PA, Visintainer PF, Shi Q, Weiss IA, Brand DA. Vitamin D and Retinopathy in Adults With Diabetes Mellitus. Arch Ophthalmol. 2012;130(6):756-60.
17. Payne JF, Ray R, Watson DG, Delille C, Rimler E, Cleveland J *et al*. Vitamin D insufficiency in diabetic retinopathy. *Endocrine Practice*.2012;18(2):185–193.
18. Suzuki A, Kotake M, Ono Y, Kato T, Oda N, Hayakawa N*et al*. Hypovitaminosis D in type 2 diabetes mellitus: association with microvascular complications and type of treatment. *Endocrine Journal*. 2006;53(4):503–510.
19. Trott M, Driscoll R, Iraldo E, Pradhan S. Associations between vitamin D status and sight threatening and non-sight threatening diabetic retinopathy:

a systematic review and meta-analysis. Journal of Diabetes & Metabolic Disorder . 2022; 21:1177–1184

20. Yau JW, Rogers SL, Kawasaki R, Lamoureux EL, Kowalski JW, Bek T *et al*. Global prevalence and major risk factors of diabetic retinopathy. Diabetes Care. 2012; 35(3):556-564.
21. Zoppini G, GallettiA, TargherG, Brangani C, Pichiri I, Trombetta M *et al*. Lower levels of 25-hydroxyvitamin D3 are associated with a higher prevalence of microvascular complications in patients with type 2 diabetes. BMJ Open Diabetes Research and Care. 2015;3(10):58.