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ORIGINAL ARTICLE

Serum Vitamin D level and its Relationship with HbA_1c in Type 2 Diabetes mellitus patients – A Comparative Study

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ABSTRACT

Background: *Diabetes mellitus is a non-communicable disease caused by multiple factors leading to insulin deficiency and insulin resistance. Vitamin D is responsible for calcium homeostasis. Vitamin D has an important role and is supposed to be a risk factor in the development & pathogenesis of type 2 diabetes mellitus by affecting either insulin sensitivity or beta cell function, or both.* ***Objectives:*** *The study was aimed to establish a relationship between serum vitamin D level and its effect on HbA_1c in type 2 diabetes mellitus patients.* ***Methods & Material:*** *This comparative study was carried out among equal number of diabetic and nondiabetic participants in the department of physiology, Rajshahi Medical college, Rajshahi. Serum vitamin D and HbA_1c was measured among the two groups. Data was analyzed by computer using the SPSS 24.* ***Result:*** *The mean BMI was 26.84 ± 3.25 & 26.79 ± 2.98 kg/m^2 in diabetic & non-diabetic group participants respectively. The serum vitamin D level was significantly ($p < 0.001$) lower in diabetic group (26.96 ± 8.54) as compared to non-diabetic group (36.51 ± 10.43). In type 2 diabetic participants a negative correlation was documented between HbA_1c and serum vitamin D level and it was statistically significant.* ***Conclusion:*** *Hypovitaminosis D was observed in type 2 diabetes mellitus patients and there was an inverse relationship between serum vitamin D and HbA_1c level.*

Keywords: *Vitamin D, HbA1c, type 2 diabetes mellitus.*

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INTRODUCTION

Diabetes mellitus is a metabolic condition marked by high blood glucose levels; it impacts not only the metabolism of carbohydrates but also disrupts the metabolism of proteins and fats, due to insufficient insulin, reduced effectiveness of insulin, or a combination of both.[1] Globally, diabetes mellitus (DM) is currently recognized as the most

prevalent metabolic noncommunicable disease, characterized by rising morbidity rates, heightened premature mortality, and escalating healthcare expenses. It is widely acknowledged that nations with low- and middle-income economies bear the greatest burden of type 2 diabetes mellitus (T2DM) on a global scale. This situation is likely attributable to the swift and ongoing socioeconomic advancements in these countries, which have led to urbanized lifestyles and the adoption of Western dietary patterns.[2] Diabetes is a metabolic disorder that has the potential to impact almost every organ system within the body. Diabetes remains a significant public health issue. It is projected that by the year 2025, approximately 380 million people globally will be affected by diabetes.[3] In 2015, Bangladesh was ranked 10th globally, with a total of 7.1 million individuals suffering from diabetes. It is projected that by 2040, approximately 13.6 million people in the country will be affected by this condition, elevating its rank to 9th at that time.[4] Diabetes mellitus is categorized into two types: Type 1 diabetes mellitus and Type 2 diabetes mellitus. Type 1 diabetes mellitus is an autoimmune disorder marked by the destruction of pancreatic β cells, resulting in a total lack of insulin in affected individuals. In contrast, Type 2 diabetes mellitus is defined by a reduction in insulin secretion and/or an increase in insulin resistance within the target tissues.[5] Given the escalating global prevalence of T2DM, there is a renewed focus on investigating the pathophysiology of this condition. The primary pathophysiological defects associated with T2DM are insulin resistance and β-cell dysfunction. This condition primarily arises from the interaction of genetic predispositions and environmental influences. The incidence of T2DM differs across various geographical areas, influenced by lifestyle choices and risk factors. In addition to traditional environmental risk factors such as obesity, lack of physical activity, consumption of high-calorie foods, and stress, the significance of specific nutritional elements in the development of T2DM is currently gaining attention.[6] Diabetes mellitus is associated with various complications like retinopathy, nephropathy, neuropathy and cardiovascular diseases and it is required to maintain adequate glycemic control, which is routinely assessed by measuring hemoglobin A_1c (HbA_1c) and fasting blood glucose levels to prevent these complications.[7] Recent research has indicated the involvement of Vitamin D in both the pathogenesis and prevention of diabetes. Numerous studies have demonstrated that Vitamin D is crucial for the proper secretion of insulin from the pancreas by activating calcium-dependent endopeptidases

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in the pancreatic β cells, while also reducing insulin resistance in target tissues, thereby enhancing glucose tolerance Elevated levels of Vitamin D have been demonstrated to reduce the occurrence of overt diabetes in populations with impaired glucose tolerance, as well as enhance glycemic control in patients with uncontrolled diabetes, according to long-term studies.[5] The role of vitamin D in glucose metabolism is thought to be linked to its function in pancreatic insulin secretion and peripheral insulin resistance. Although it is categorized as a vitamin, vitamin D is also recognized for its multiple hormonal roles, which are believed to arise from its interaction with vitamin D receptors (VDRs) that are extensively present on different cell types. One of these roles includes the effect of vitamin D on the VDRs located in pancreatic beta islet cells.[7] The discovery of vitamin D receptor in tissues which are not involved in calcium and bone metabolism like pancreatic beta cell and immune cells broadens the physiological role of vitamin D. Numerous animal studies have demonstrated that vitamin D is crucial for the proper secretion of insulin from the pancreas. A variety of studies have indicated its significance in enhancing insulin production and secretion in humans, while also reducing insulin resistance. [3] Globally, vitamin D deficiency is a prevalent issue within the general population. Despite the fact that vitamin D deficiency is typically unexpected in tropical regions such as the Indian subcontinent, numerous cross-sectional studies have revealed a notably high prevalence of vitamin D deficiency, even among healthy individuals from various subsets of this population. Recent research has indicated a significantly high occurrence of suboptimal vitamin D levels in patients with Type 2 diabetes mellitus in this subcontinent; however, some of these studies have noted that the prevalence was not significantly elevated when compared to that of normal individuals. [4] As there was limited data on relationship between vitamin D levels and glycosylated hemoglobin (HbA_1c) levels in Type 2 diabetes mellitus patients especially in Bangladesh. Keeping this in mind, this study was conducted with the objective to determine the serum vitamin D level in Type 2 diabetes mellitus patients and to establish a relationship between serum vitamin D level and HbA_1c level.

METHODS & MATERIALS

Study place & study subjects

This cross-sectional analytical study was done in the Department of Physiology, Rajshahi Medical College, Rajshahi from 1st January 2022 to 31th December 2022. According to selection criteria 42 Type 2 diabetes mellitus patients who were on oral anti diabetic drug for at least 6 months were selected from outpatient department of Rajshahi diabetic Association hospital and they were enrolled as Group A. Another 42 apparently healthy individual from staff of the hospital, relatives of patients and other volunteers were enrolled as Group B. The method of sample collection was purposive sampling.

Ethical Considerations

Approval was obtained from the Ethical Review Committee of Rajshahi Medical College. After enrolment informed written consent was taken from the participants.

Blood Collection & Laboratory Investigations

With full aseptic precautions (about 10 ml) was collected from anterior cubital fossa by venipuncture technique using a 21-gauge hypodermic needle and was collected in a sterile container. 2 ml blood was mixed with EDTA for estimation of HbA1c. The remaining blood was allowed to clot and there after centrifuged at 2000 RPM for 5 min at room temperature (29° C - 31° C). Serum vitamin D was measured by enzyme linked immunoassay (ELISA) was designed by Calbiotech, Inc. and HbA_1c was measured by colorimetric determination of glycated hemoglobin in whole blood using Stanbio Kit, Texas-USA.

Data Analysis

After collection of all the required data, these were checked, verified for consistency and then tabulated into the computer using the Package for Social Sciences (SPSS version 24). Statistical analysis was carried out using appropriate statistical equation. Continuous variables were expressed as mean±SD and median whereas qualitative or categorical variables were described as frequencies and proportions. An independent t-test was used to compare continuous variables with two categories and ANOVA test was used to compare continuous variables more than two categories. A Chi square test was used to compare categorical variables with two categories. Correlation of serum vitamin D and HbA1c was determined by Pearson's correlation analysis. A p-value of <0.05 was considered statistically significant.

RESULTS

Out of a total of 84 participants enrolled for the study, 50 were males and 34 were females. The mean age of Group A participants was 50.57 ± 6.86 years and that of the Group B participants was 48.93 ± 5.04 years. In this study the mean BMI was 26.84 ± 3.25 and 26.79 ± 2.98 kg/m2 of Group A and Group B participants respectively. In diabetic group, 64.30% of the patients were overweight, 23.80% had normal BMI and remaining 11.90% were obese. On the other hand, in the non-diabetic group, 47.60% of the respondents were overweight, 35.70% had normal BMI and only 16.70% were obese (*Table I*).

Table – I: Distribution of the respondents according to BMI

BMI (kg/m^2)	Group A (n=42)	Group B (n=42)
Frequency (%)		
Normal (18.5 to 24.9)	10 (23.80%)	15 (35.70%)
Overweight (25 to 29.9)	27 (64.30%)	20 (47.60%)
Obese (30 to 39.9)	5 (11.90%)	7 (16.70%)
Total	42 (100.00%)	42 (100.00%)
(mean ± SD)	26.84 ± 3.25	26.79 ± 2.98

Table II shows that among the diabetic patients 57.10% had vitamin D insufficiency, 23.80% had vitamin D sufficiency and remaining 19.00% had vitamin D deficiency. In the non-diabetic participants, 76.20% had sufficient vitamin D, 14.30% insufficient and only 9.50% had deficient serum vitamin D level.

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Table – II: Serum vitamin D level in Group A and Group B Participants

Serum vitamin D(ng/ml)	Group A (n=42)	Group B (n=42)
Frequency (%)		
Deficient (< 20 ng/ml)	8 (19.00%)	4 (9.50%)
Insufficient (20-29 ng/ml)	24 (57.10%)	6 (14.30%)
Sufficient (30-100 ng/ml)	10 (23.80%)	32 (76.20%)
Total	42 (100.00%)	42 (100.00%)

In diabetic patients, 83.30% of the patients had $HbA_1c \geq 6.5\%$ and remaining 16.70% had < 6.5%. On the other hand, in the non-diabetic participants, all (100.00%) had $HbA_1c < 6.5\%$ (*Table III*).

Table – III: HbA_1c level in Group A and Group B Participants

HbA_1c level (%)	Group A (n=42)	Group B (n=42)
	Frequency (%)	
< 6.5	7 (16.70%)	42 (100.00%)
≥ 6.5	35 (83.30%)	0 (0.00%)
Total	42 (100.00%)	42 (100.00%)

Table IV shows no statistically significant difference of serum vitamin D level among the different duration of diabetes mellitus patients (p > 0.05).

Table – IV: Relationship between duration of diabetes mellitus and serum vitamin D level of the diabetic patients (*n*=42)

Duration of diabetes (year)	Vitamin D level (ng/ml)	F, p value
< 1 year	30.69 ± 16.03	0.41, > 0.05
1-5 years	26.65 ± 7.53	
> 5 years	26.44 ± 8.03	

During the comparison of serum vitamin D level between the diabetic and non-diabetic group, the mean serum vitamin D level of diabetic group (26.96 ± 8.54 ng/ml) was significantly lower (p < 0.001) than that of the non-diabetic group (36.51 ± 10.43 ng/ml) (*Table V*).

Table – V: Comparison of serum vitamin D level between Group A and Group B Participants

Group	Serum vitamin D #		t-value	p-value
	mean ± SD (ng/ml)	Range (ng/ml)		
Group A (n = 42)	26.96 ± 8.54	11.67 to 53.94	4.59	< 0.001
Group B (n = 42)	36.51 ± 10.43	13.84 to 58.29		

(# Data were analyzed by Unpaired t-Test and were presented as mean ± SD.)

Table VI showing that mean HbA_1c level of diabetic group (7.24 ± 0.77%) was significantly (p < 0.001) higher than that of non diabetic group (5.20 ± 0.31%). In *Figure 1* scatter diagram showing significant (p<0.05) negative correlation between HbA_1c and serum vitamin D level in both diabetic group (r= - 0.31) and nondiabetic group (r= - 0.45).

Table – VI: Comparison of HbA_1c between Group A and Group B Participants

Group	HbA1c #		t-value	p-value
	mean ± SD (%)	Range (%)		
Group A (n = 42)	7.24 ± 0.77	5.60 to 8.70	15.96	< 0.001
Group B (n = 42)	5.20 ± 0.31	4.60 to 5.70		

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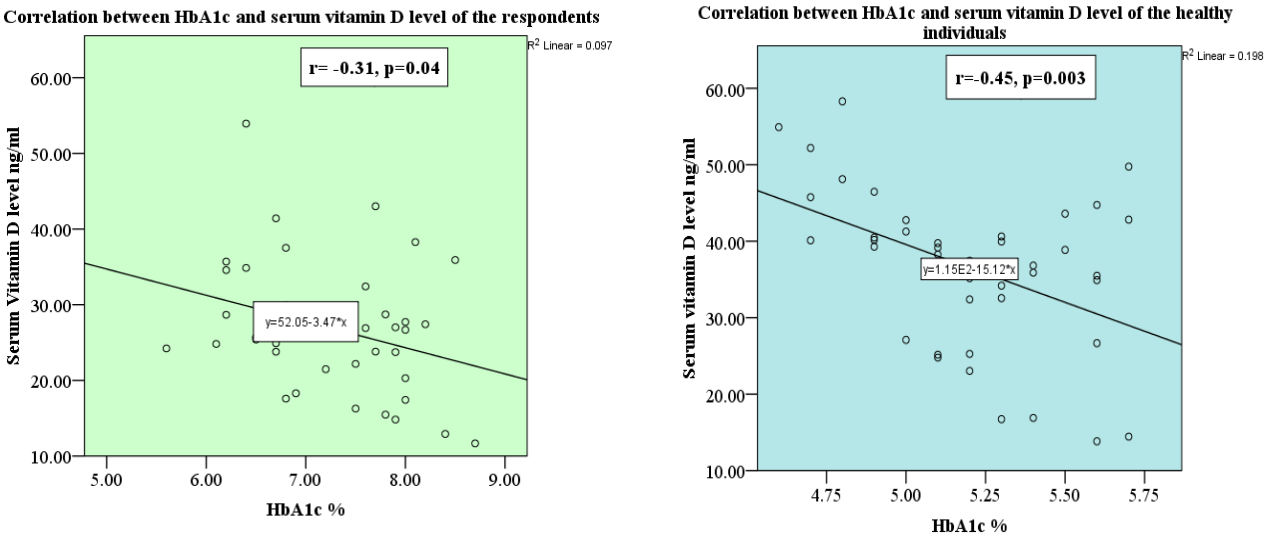

Figure – 1: Scatter diagram showing correlation between HbA₁c and serum vitamin D level in diabetic and non-diabetic group (*n*=42 in each group)

DISCUSSION

The present study was under taken to assess serum vitamin D level and its relationship with HbA1c in type 2 diabetes mellitus patients. A total of 84 study subjects (42 in diabetic group and 42 in non-diabetic group) were involved in the study after taking informed written consent data were collected through a semi-structured questionnaire. In the present study the distribution of the respondents according to their BMI revealed that in diabetic group, 64.30% of the patients were overweight, 23.80% had BMI within the normal range and remaining 11.90% were obese. On the other hand, in the non-diabetic group, 47.60% respondents were overweight, 35.70% had BMI within the normal range and 16.70% were obese. Obese respondents were higher in non-diabetic group (16.70%) than the diabetic group (11.90%). The mean BMI of the diabetic patients was 26.84 ± 3.25 kg/m2 and that of the non-diabetic participants was 26.79 ± 2.98 kg/m2. Similar observations were made by many researchers of different countries.[8] Findings of this present study were not in agreement with studies done by different researcher. [9,10] The dissimilarities might be due to the studies were done in different geographical region and variation in lifestyle. In the current study among the diabetic patients 57.10% were in vitamin D insufficiency state, 23.80% were in sufficiency state and remaining 19.00% were in deficiency state. On the other hand, in the non-diabetic group, 76.20% of the respondents were in vitamin D sufficiency state, 14.30% were in insufficiency state and only 9.50% were in deficiency state. Similar observations were made by many researchers of different countries.[6,11,12] Result of this study was not similar to some studies done by many researchers.[3,13,14] Findings conflicting to the present study were also found in a study done in Saudi Arabia where in diabetic group 35.60% patients had vitamin D deficiency, 19.60% had vitamin D insufficiency, 44.70% had vitamin D sufficiency and in healthy control group all of the respondents were in vitamin D sufficiency state.[10] Contradictory findings also found with the studies done by other researchers.[5,15,16] In this study, glycemic status of the respondents showed that 83.30% of the patients had HbA₁c ≥ 6.5% and remaining 16.70% had < 6.5% in diabetic group. On the other hand, in the non-diabetic group, all (100.00%) of the respondents had HbA₁c < 6.5%. This result was almost similar to another study.[5] In the current study, serum vitamin D level in diabetic group ($\bar{x}$=26.96, SD=8.54) was significantly lower (mean difference 9.55, 95% CI [5.41, 13.69]) than the non-diabetic group ($\bar{x}$=36.51, SD=10.43) and it was statistically significant (p < 0.001). This finding was in agreement with other studies. [10,13] In this study, HbA1c level in diabetic group ($\bar{x}$= 7.24, SD = 0.77) was significantly higher (mean difference 2.04, 95% CI [1.78, 2.29] than the non-diabetic group ($\bar{x}$=5.20, SD=0.31) and it was statistically significant (p < 0.001). Similar result was found in other studies.[10] Scatter diagram revealed negative correlation (r = - 0.31) between HbA₁c and serum vitamin D level in diabetic group and it was statistically significant (p=0.04). This finding was in agreement with other studies. [3,5,6,17]

CONCLUSION

From the results of this study, it can be concluded that the type 2 diabetic patients had a lower level of serum vitamin D than the healthy adults and serum vitamin D level is negatively correlated with their HbA1c level.

CONFLICT OF INTEREST

There is no conflict of interest among the authors.

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