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Association of Serum Vitamin D_3 and Glycated Haemoglobin in Type II Diabetes Mellitus Patients

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Abstract: Type 2 Diabetes Mellitus is a major health concern globally. The total number of Diabetics is expected to reach 366 million by 2030. Efforts should also be made in preventing Diabetes Mellitus. Diabetes Mellitus, cancer, multiple sclerosis, psoriasis, cardiovascular diseases and metabolic syndrome are some of the non-skeletal diseases where Vitamin D_3 deficiency has been implicated to play a vital role. Many studies indicate that calcium and Vitamin D_3 homeostasis play an important role in development of Diabetes Mellitus. The Present study was carried out to evaluate the association between the levels of Vitamin D_3 and Glycated Haemoglobin (HbA1c) in Type 2 diabetes mellitus. An Observational study was conducted at Shri Krishna Hospital, Karamsad between 100 cases of type 2 diabetes mellitus and 100 normal healthy controls. HbA1c and Renal Function was measured in both groups by validated instruments. Correlation between HbA1c and Vitamin D_3 was calculated. In the study, Vitamin D_3 deficiency was observed in 54% of cases and 68% of the controls. Mean Vitamin D_3 levels were also significantly deficient in control group (p-0.02). There was a significant negative correlation between fasting blood glucose and vitamin D_3 level. It concludes that in diabetic patients, there was a negative correlation between HbA1c and Vitamin D_3 levels pointing that probably, Vitamin D_3 deficiency as one of the cause in the possession of diabetes mellitus

Keywords: Vitamin D_3, Glycated Haemoglobin, type 2 Diabetes Mellitus.

INTRODUCTION

Type 2 diabetes mellitus (T2DM) is a group of metabolic disorders characterized by hyperglycaemia. Several different types of DM are caused by complex interactions of genetic and environmental factors. Depending on the etiology of DM, factors which contribute to hyperglycaemia include reduced insulin secretion, insulin résistance, and increased glucose production [1]. In 2017, International Diabetes Federation (IDF) estimated that 425 million people have diabetes which is expected to rise to 629 million by the year 2045.The greatest number of people with diabetes fall between 40 to 60 years of age. In India, alone about 73 million people are living with diabetes. So this places India second to China. At present in India, 8.8 % population is diabetic and it is expected to rise to 10.2% (123 million) by the year 2035. It significantly increases the risk of cardiovascular diseases and stroke, Although insulin resistance and beta cell decompensation compose the well accepted basis of type 2 diabetes mellitus, the molecular mechanism underlying their development are incompletely understood [2]. Vitamin D_3 supplementation to prevent Diabetes Mellitus has been explored. The primary route via which people obtain vitamin D_3 is through exposure to ultraviolet B (UVB) rays of sunlight at wavelengths between 290-315 nm [3]. Recently, the extra skeletal effects of vitamin D_3 have raised considerable interest. Vitamin D_3 deficiency appears to be related to the development of diabetes mellitus type 2. Mild to moderate vitamin D insufficiency has been proposed as a risk factor for type 2 diabetes. Higher plasma vitamin D_3 levels have been shown to be associated with a lower risk for the development of diabetes mellitus in high risk patients [4]. Several studies have demonstrated the role of vitamin D_3 in the regulation of the endocrine function of pancreas especially the beta cells. Despite the fact that insulin secretion and action being the cause of diabetes, in routine clinical care, insulin is rarely measured, even though it can be measured easily on automated equipment in laboratories. Blood glucose control in diabetic patients is usually monitored by determination of HbA1c levels (glycated haemoglobin)

which is now the gold standard, and gives an estimate of the amount of glucose in the blood over the previous three months [5].

To the best of my knowledge, studies on the role of vitamin D3 deficiency in diabetics in Gujarat are few. Therefore, this study is undertaken. It aims to provide a comprehensive and comparative study of vitamin D3 and glycemic control parameters in patients of Type 2 DM. This would help to determine the correlation of HbA1c and Vitamin D3 in Type-2 Diabetes Mellitus patients.

MATERIALS AND METHODS

Study type, Study setting and Study Period

A hospital based case control study was carried out in Shri Krishna Hospital in Karamsad city of Gujarat from December 2014 to November 2015.

Participants recruitment procedure

100 participants in the study group and 100 participants in the control groups were enrolled by the following procedure.

Selection of study group

Persons aged 30 years or more who came to hospital for routine health check-up were checked for eligibility criteria. Routine health check-up scheme includes basic information, clinical history and laboratory investigations. (FPG, HbA1c, RFT, Vitamin D_3 etc.)

Inclusion Criteria

Known and newly diagnosed Diabetic patients with FPG >126 mg/dl ,HbA1c > 6.5%, Serum Urea < 40 mg/dl , Serum Creatinine < 1.3mg/dl were included in the study.

Exclusion Criteria

Non Diabetic individuals, individuals on Vitamin D_3/calcium Supplements and individuals with Liver or kidney diseases were excluded from the study.

Selection of control group

Inclusion Criteria

Age and sex matched normal healthy individuals with FPG <110 mg/dl, HbA1c< 6.5%, Serum Urea < 40 mg/dl, Serum Creatinine <1.3mg/dl.

Exclusion Criteria

Individuals on Vitamin D_3/calcium Supplements and individuals with Liver or kidney diseases were excluded from the study.

Blood sample collection and processing

Samples were collected with an aseptic blood collection technique with the use of sterile gloves and disinfection of venepuncture site with 70% ethyl alcohol. All the samples were collected in sitting position. Blood samples were collected in three vacutainers: Plain bulb for Renal function tests and Vitamin D3, Sodium fluoride bulb for Fasting plasma glucose and EDTA tube for HbA1c. Samples were centrifuged at 1500 rpm for 15 minutes within one hour of collection to obtain serum/plasma. These were processed in the biochemistry laboratory for the estimation of FPG, HbA1c, Vitamin D, RFT.

Estimation of Plasma glucose [6]

Plasma glucose was estimated by Hexokinase method in in fully automated Roche Cobas Integra 400 plus clinical chemistry analyser.

Estimation of glycated haemoglobin (HbA1c) [7]

HbA1c was measured by Immunoturbidimetry Standardized according to IFCC method in fully automated Roche Cobas Integra 400 plus clinical chemistry analyser.

Vitamin D_3 estimation [8]

Vitamin D_3 was measured by electrochemiluminescence (ECL) method in Roche Cobas E - 411 Immunoassay Analyser. Vitamin D_3 level less than 50 nmol/Litre was considered as deficiency.

RFT was estimated in fully automated Roche Cobas Integra 400 plus clinical chemistry analyser.

Statistical analysis

Data were entered and analysed through Epi info 7. Categorical variables were expressed through percentages while continuous variables were expressed as mean and standard deviation. Spearman correlation coefficient were calculated to know the relation between correlation between Vitamin D_3 levels and Glycated haemoglobin in cases of Type 2 Diabetes Mellitus. A p-value less than 0.05 was considered as statistically significant.

RESULTS

Table-1: Characteristics of the participants

Variables	Case (n=100)	Controls (n=100)	P value*
Age (Years)	55.09 ± 11.03	52.54 ± 9.06	0.06
Gender Female Male	 42 (46.67%) 58 (52.73%)	 48(53.33%) 52(47.27%)	0.48
FPG (mg/dl)	165.92 ± 45.86	94.63 ± 9.20	<0.0001
HbA1c (%)	8.00 ± 1.58	5.65 ± 0.53	<0.0001
Vitamin D_3 (nmol/L)	55.82 ± 33.33	42.78 ± 27.21	0.02
Vitamin D_3 (nmol/L) <50(Deficiency) ≥50 (sufficiency)	 54 (54%) 46 (46%)	 68 (68%) 32 (32%)	0.059

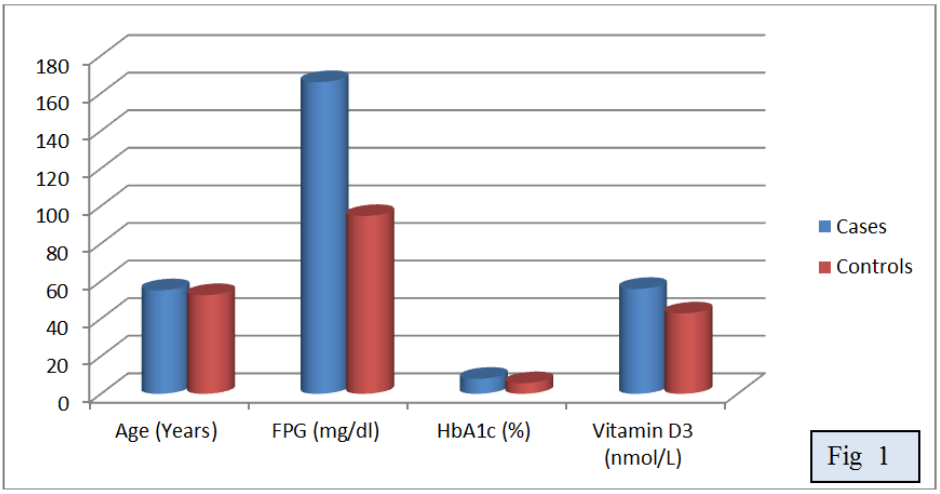

Fig-1: Characteristics of the participants

Table-1& Fig-1 shows that mean age among control groups was slightly lower than the mean age of the case group. Fasting plasma glucose and Glycosylated haemoglobin were significantly lower in control group than study group (p value <0.0001 in both). Mean Vitamin D_3 level were also significantly deficient in 54% of cases and 68% controls, which is statistically insignificant.

Table-2: Vitamin D_3 levels according to HbA1c levels in Cases with T2 DM

Vitamin D_3 (nmol/L)	HbA1c <7.5%	HbA1c >7.5%	Total T2 DM Cases
<50 (Deficiency)	25	29	54
≥50 (Sufficiency)	26	20	46
TOTAL	51	49	100

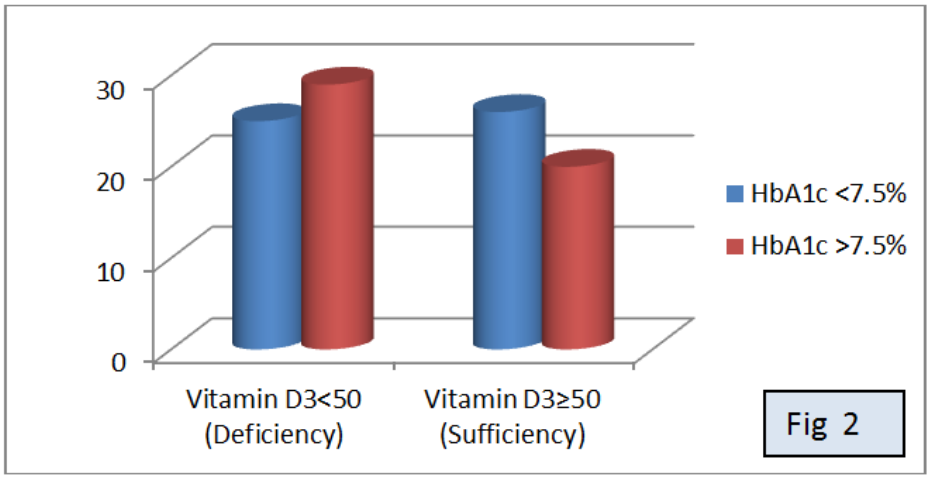

Fig-2: Vitamin D_3 levels according to HbA1c levels in Cases with T2 DM

Table-2 & Fig-2 shows Categorical comparison of vitamin D_3 deficiency and glycemic control having HbA1c < 7.5 % and > < 7.5 %.. Bar Graph also suggests that the tallest bar is representing vitamin D levels less than 50 and HbA1c levels of more than 7.5 %. This also establishes that the correlation present between both these parameters is an inverse correlation. (p value 0.412).

Table-3: Correlation of vitamin D_3 with HbA1c

Parameters	Pearson's Correlation Coefficient (Vitamin D_3WITH VIT-D_3)	p Value
HbA1c-Vitamin D_3	- 0.2767	0.005

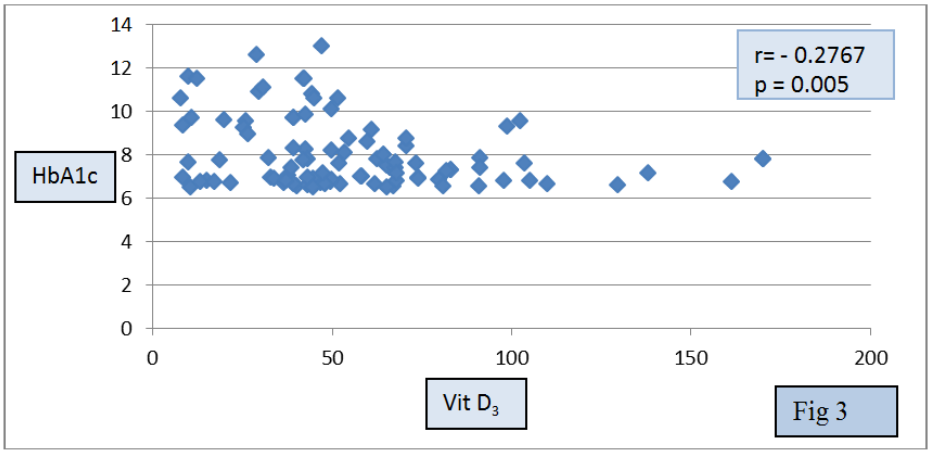

Table-3 shows Correlation of vitamin D_3 with HbA1c & Fig-3 shows scattered graph showing correlation of vitamin D_3 with HbA1c. There was significantly negative or inverse correlation between HbA1c level and vitamin D_3 level (correlation coefficient is -0.2767, p value 0.005) among study participants.

DISCUSSION

This study was a case control study, done over a period of one year from December 2014 to November 2015. It included 100 cases of Type 2 Diabetes Mellitus who had FBS and HbA1c levels >126 mg/dl and >6.5% respectively. All the controls had normal HbA1c (5.4-6.4%) and FBS (70-110mg/dl) levels. The RFT was normal in both the groups with S. creatinine (<1.3mg/dl) and S. urea (<40mg/dl). There was significantly negative or inverse correlation between HbA1c level and vitamin D_3 level (correlation coefficient is -0.2767, p value 0.005) among study participants. In present study, Vitamin D_3 deficiency was observed in 54% of cases and 68% of control. A study done by AK SV *et al.*, found that Vitamin D deficiency was observed in 32% of cases and 25% of controls while Sheth *et al.*, [9] observed Vitamin D_3 deficiency in 91.4% of cases of Type 2 Diabetes Mellitus and 93% in the control group. In a cross sectional Iranian study by Taheri and colleagues [10], the prevalence of vitamin D_3 deficiency was 83.3% in diabetic patients and 75.6% in healthy subjects. Another

cross sectional study among rural and urban adult Indians, Harinarayan *et al*., [11] also observed 44% and 62% for rural and urban men respectively and 70% and 75% deficiency for rural and urban women respectively. High deficiency in the present study participants may be due to life style, underlying health condition, poor exposure to sunlight, poor diet or increase age.

In our study, there was significantly negative correlation between HbA1c and vitamin D_3 level. Study done by Ahmadieh H *et al*., [12] and Giacomo Zoppini *et al*., [13] also showed negative association between vitamin D_3 level and glycemic control. While Sheth J *et al*., [9] studied that there was no association of serum Vitamin D_3 deficiency on HbA1c. Chiu *et al*., [14] found that vitamin D_3 deficiency was related to a higher risk for insulin resistance and the metabolic syndrome. There are several lines of evidence to support that vitamin D_3 influences impaired β-cell function, insulin resistance, and systematic inflammation [15]. It has been demonstrated that vitamin D_3 receptors exist in many tissues including pancreatic β-cells [16], allowing vitamin D_3 to potentially modulate the insulin response to elevated blood glucose.

CONCLUSION

The present study revealed that the patients with Type 2 DM have a deficiency of Vitamin D_3 levels but the controls also have deficient vitamin D_3 levels surprisingly which may be due to lack of exposure to sunlight. Vitamin D_3 levels are usually not estimated in routine practice along with other investigations for follow up in Type 2 DM patients as, they do no not have any signs and symptoms of underlying deficiency, so it remains hidden and most of the times undiagnosed. Therefore, for the early diagnosis of Vitamin D_3 deficiency, its estimation is required in Type 2 Diabetes mellitus patients because vitamin D_3 levels have a protective role in diabetes mellitus type 2. Vitamin D_3 supplementation should be considered in patients with type 2 diabetes mellitus, it may help to improve the glycemic control. Vitamin D_3 fortified diabetic diets may be of additional help.

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