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Vitamin D Deficiency in Type II Diabetic Patients in Jordan: A Retrospective Longitudinal Study

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ABSTRACT

Background: Previous research suggested that vitamin D is linked to glycemic control, insulin resistance, and Type 2 Diabetes. Vitamin D regulates insulin secretion, and its deficiency can decrease insulin sensitivity and increase insulin resistance.

Aims: The research aimed to determine whether vitamin D levels could significantly affect their glycemic control, as reflected in the changes in their fasting blood glucose (FBG) and glycosylated hemoglobin (HbA1c) levels.

Methods: At the Prince Hamza Hospital and Diabetes Center, in Amman, Jordan, a study was conducted on 392 diabetic patients. These patients were retrospectively evaluated for their last three readings of serum vitamin D, calcium, and phosphate, and other relevant data which are performed on their routine follow-up.

Results: The study population consisted of 201 females (53.1%) and 191 males (48.7%), who had a median age of 57 (IQR: 8) and a BMI of 32 (IQR: 7.28). The analysis found that the controlled and poorly controlled groups had a significant difference in vitamin D levels (P = 0.036). Additionally, the analysis showed that as serum vitamin D levels increased over nine months, HbA1c levels decreased, leading to better diabetes control. Furthermore, the study found that the mean difference of vitamin D could significantly predict the mean difference of both FBG (P=0.033) and HbA1c (P<0.001).

Conclusion: Vitamin D deficiency is linked to poor glycemic control in Jordanian patients, and their serum vitamin D levels can predict glycemic status. However, further studies are needed to analyze this association due to confounding factors.

Keywords: Glycemic Control; Vitamin D Deficiency; Type II Diabetes; Insulin Resistance; Jordan; Retrospective

Abbreviations: FBG: Fasting Blood Glucose; T2DM: Type 2 Diabetes Mellitus; NIDDM: Non-Insulin Dependent Diabetes Mellitus; MENA: Middle East and North Africa; IDF: International Diabetes Federation; SD: Standard Deviation; DCCT: Diabetes Control and Complications Trial

Introduction

Diabetes mellitus (DM) is a chronic metabolic disorder of various causes in which the body cannot metabolize carbohydrates, fats, and proteins due to defects in insulin excretion and functioning [1]. It is considered the seventh leading cause of death worldwide [2]. Type 2 diabetes mellitus (T2DM), also known as non-insulin dependent diabetes mellitus (NIDDM), is caused by insensitivity of the cells towards insulin. Insulin is not able to stimulate glucose uptake in peripheral target organs such as muscle and fat leading to insulin resistance, glucose intolerance, and higher insulin levels in blood than normal [3]. The incidence and prevalence of type 2 DM is increasing at an alarming rate all over the world [4]. Middle East and North Africa (MENA) had the highest global prevalence of diabetes in 2011, according to the International Diabetes Federation (IDF) (12.5 percent). By the year 2030, 14.3 percent is projected to have been

achieved [5]. Vitamin D is considered one of the important human nutrients that can be obtained endogenously and exogenously [6]. The main source of vitamin D is synthesized endogenously through the skin with the help of sunlight irradiation, with limited dietary sources [7]. The most important and well-known function of vitamin D is to maintain calcium and phosphorus homeostasis and support bone health [4].

Recently, there is increasing evidence that alterations in vitamin D and calcium are associated with the development of type 2 diabetes [8]. Vitamin D is involved in the regulation of insulin secretion where its receptors are present in the pancreatic β cells [9]. There is an inverse relationship between Vitamin D levels and HbA1C, low Vitamin D Levels are related to increased incidence of T2DM [9]. It is stated that a vitamin D deficiency was associated with an accelerated occurrence of metabolic syndrome. Therefore, reduced vitamin D levels have been related to reduced insulin sensitivity, increased insulin resistance, and high fast blood glucose [4]. Previous studies proved that the treatment of vitamin D deficiency among type 2 diabetic patients can improve their laboratory results [10]. Research has confirmed that maintaining adequate levels of vitamin D can promote insulin sensitivity while lacking this vital nutrient can impair pancreatic beta cell function. Furthermore, this study suggests that vitamin D deficiency may significantly increase the risk of insulin resistance, type 2 diabetes, and metabolic syndrome, regardless of other contributing factors [10]. A study in Saudi Arabia concluded that vitamin D deficiency was remarkably higher in T2DM patients than the non-diabetic individuals and the only factor that positively correlated with vitamin D deficiency was the duration of diabetes. Therefore, early screening and treatment of vitamin D deficiency is recommended. Vitamin D must be added for individuals with high-risk features for diabetes, with prediabetic patients to help prevent the incidence of the disease [4]. To the best of our knowledge, no previous study was carried out in Jordan to determine the possible association between vitamin D deficiency and the status of glycemic control among diabetic patients. Accordingly, this study was designed to determine the association between vitamin D deficiency and the status of glycemic control among diabetic patients and to study the predictability between vitamin D level and the possible glycemic control.

Methods

Study Design and Settings

This study was conducted with ethical approval from the institutional review board (IRB) of the Hashemite University and Prince Hamza Hospital. Approval was also obtained to access hospital records. The authors were responsible for the study›s design, data collection and analysis, and the preparation of the manuscript. They ensure the accuracy, completeness of the data and the study›s fidelity to the approved protocol. The study enrolled 392 participants over a period of nine months.

Participants

This is a retrospective longitudinal study, where the authors utilized data from electrical patient's records of the Diabetes Center at Prince Hamza Hospital in Amman, Jordan. The study involved 392 records for adult patients between 18 and 65 years old who had a laboratory-evidenced diagnosis of Type II diabetes and were being treated with diabetes, over a period of nine months. The criteria for diagnosing diabetes included fasting plasma glucose levels (FBG) greater than or equal to 126 mg/dl (7 mmol/L) and HbA1c (glycated hemoglobin) levels greater than or equal to 6.5. Patients› height and weight were measured while they were wearing light clothes and after taking off their shoes, and their BMI (Body Mass Index) as the ratio of weight in kilograms to the square of height in meters. The BMI was classified according to WHO criteria as normal (< 25), overweight (25 - 29.9), and obese class I (30 -34.9), obesity class II (35 – 39.9), and obesity class III (>= 40). Finally, the patients› PTH (parathyroid hormone) levels were measured and considered normal (9-55 pg/mL), or high (55 pg/mL).

The inclusion criteria were:

1) Patients aged 18–65 years old,

2) Diagnosed with T2DM >3 months ago,

3) Followed up regarding DM on a regular basis at the Prince Hamza Diabetes and Endocrine Center within the same period (March-October, 2023).

The exclusion criteria were:

1) Patients aged <18 years and >65,

2) Those who were recently diagnosed with T2DM,

3) Patients diagnosed with DM type I,

4) Pregnant women, and

5) Patients on vitamin D supplements or drugs that affect vitamin D metabolism such as corticosteroids.

To maintain high consistency of the results, the authors only considered patients who had their laboratory tests carried out for the same period from March 2023 to October 2023 with 3-months interval in between. We collected data that was documented on the system for the intended outcomes three times, each within a three-month interval. The first reading was taken in March 2023, followed by the second reading in July 2023, and the third reading in October 2023.

In accordance with the ADA guidelines [11], the authors have categorized the patients based on their average HbA1c readings to better reflect their diabetic control. The patients were divided into two groups - controlled diabetes (HbA1c < 7.5 mmol/mol) and poorly controlled diabetes (HbA1c > 7.5 mmol/mol). Similarly, the patients were also classified based on their FBG readings - controlled (< 7.2

mmol/L) and poorly controlled (> 7.2 mmol/L) DM. Furthermore, we categorized the patients based on their vitamin D levels, where patients with < 20ng/dl were considered deficient, > 20ng/dl and < 100ng/dl were insufficient, and > 100ng/dl were sufficient. As all the patients were either deficient or insufficient in vitamin D, only these two groups were illustrated in our study. Regarding serum calcium level, patients with levels between 2.11 and 2.7 were considered as normal, whereas less than 2.11 was considered hypocalcemia, and more than 2.7 was hypercalcemia. Additionally, phosphate levels > 0.81 and <1.45 were deemed normal, < 0.81 was considered hypophosphatemia, and >1.45 as hyperphosphatemia.

Outcome Measures

The study hypothesized that patients who had vitamin D deficiency (<20 ng/dL) or insufficiency (>20 but < 100 ng/dL) had a higher chance of having poor glycemic control (defined as patients who had average blood glucose measurements on three consecutive visits > 130 or < 70 mg/dL) [10]. Moreover, the hypothesis also suggested that diabetic patients who had improving serum vitamin D levels over time, would have decreasing levels of FBG and HbA1c, thus, better glycemic control. Accordingly, at each point of time we obtained FBG, HbA1c, vitamin D levels, in addition to blood pressure that were measured and documented on each visit every 3 months for each patient. Other laboratory results including PTH, serum calcium, phosphate, weight, height, and BMI were obtained on the second visit, on July 2023. The authors intended to obtain the levels of serum calcium and phosphate due to the evidence that serum vitamin D levels has a direct effect on the levels of these minerals, and therefore they could have a role in the glycemic control in diabetic patients.

Statistical Analysis

The data analysis was conducted using Jamovi 2.3.17 and RStudio. Kruskal-Wallis test was performed to investigate if there is significant difference between the patients regarding the demographic characteristics and other laboratory readings. Normally distributed data were illustrated as Mean and standard deviation (SD), non-normally distributed data were illustrated as median and interquartile range. The average of the three readings of HbA1c, FBG, and vitamin D was calculated and accordingly categorized the patients into the previously mentioned categories. The authors then studied the association between vitamin D levels and glycemic control using Chi-Square test. Additionally, to investigate if there is a significant difference in the level of vitamin D among controlled and poorly controlled diabetic patients, one-way ANOVA test was performed. In order to investigate if there is a relevant correlation between FBG, HbA1, vitamin D levels, serum calcium, and phosphate, we calculated the mean difference of the three readings taken for each variable and then measured the correlation between the mean differences and the correlation between these differences and serum calcium and phosphate levels, using Spearman correlation. Moreover, we performed linear regression analyses to determine changes over time for the intended outcome measures.

Results

Characteristics of the Population

The study included 392 diabetic patients; 201 females (53.1%) and 191 males (48.7%), with a median age of 57 (IQR: 8) and a BMI of 32 (IQR: 7.28). On every 3-month point of time, a reading of fasting blood glucose (FBG), HbA1c, vitamin D, and systolic and diastolic blood pressure (BP) were collected. All data are demonstrated in Table 1 (Figure 1).

Table 1: The sociodemographic and the relevant data collected for the included patients.

	Patients (n=392) Median (IQR)
Age	57.0 (8)
Height	166 (11)
Weight	88.0 (20)
BMI (kg/m^2)	32.0 (7.28)
Fasting blood glucose 1st reading	10.32 (5.455)
Fasting blood glucose 2nd reading	10.82 (6.420)
Fasting blood glucose 3rd reading	11.29 (6.820)
Average fasting blood glucose (mmol/l)	11.25 (5.38)
HbA1c 1st reading (mmol/mol)	8.50 (2.2)
HbA1c 2nd reading (mmol/mol)	8.70 (2.725)
HbA1c 3rd reading (mmol/mol)	9.00 (2.7)
Average HbA1c (mmol/mol)	8.83 (2.2)
Vitamin D 1st reading (ng/ml)	19.84 (12.85)
Vitamin D 2nd reading (ng/ml)	17.25 (13.55)
Vitamin D 3rd reading (ng/ml)	15.05 (12.275)
Average vitamin D (ng/ml)	18.60 (10.684)
Calcium level (mmol/l)	2.35 (0.192)
Phosphate level (mmol/l)	1.13 (0.260)
Systolic BP 1st reading (mmHg)	136.00 (28)
Diastolic BP1st reading (mmHg)	79.50 (15.25)
Systolic BP 2nd reading (mmHg)	136 (26)
Diastolic BP 2nd reading (mmHg) (mean & SD)	80 (10.55)
Systolic BP 3rd reading (mmhg)	133 (27.25)
Diastolic BP 3rd reading (mmhg)	80 (15)
Average systolic BP (mmhg)	135.00 (23.25)
Average diastolic BP (mmhg)	79.00 (11)

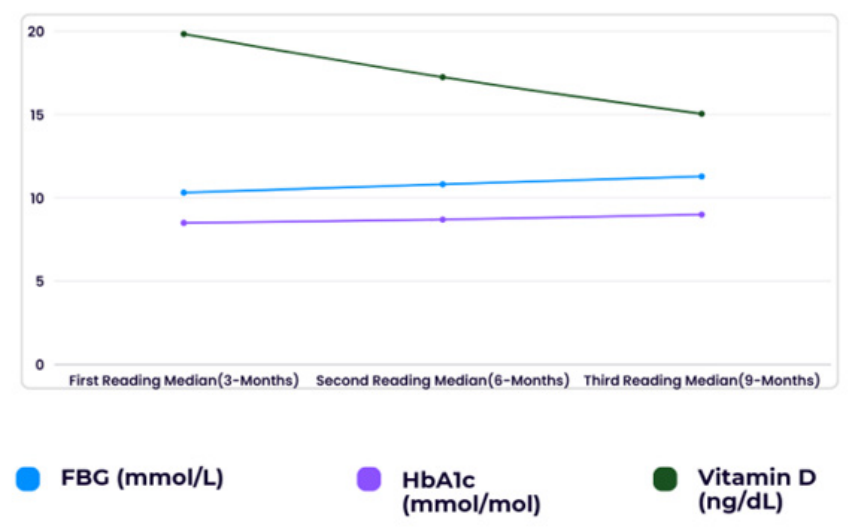

Figure 1: The time trend of FBG, HbA1c, and Vitamin D median readings over the nine months.

The Association Between HbA1c and Vitamin D Groups

The authors used the previously mentioned categories in order to study the association between HbA1c levels which reflect the previous control of diabetes, with vitamin D levels. According to Table 2, the majority of patients with vitamin D deficiency also had uncontrolled diabetes (93.4%), and only 6.6% of vitamin D-deficient patients having controlled diabetes. Moreover, the poorly-controlled group consisted the most of the insufficient vitamin D group (83%), with a P value of 0.001. Such finding underlines the significance of vitamin D deficiency among patients with uncontrolled diabetes. Although the other findings were not statistically significant (P=0.284 and P=0.09), and the majority of patients, regardless of their glycemic control status, had normal serum levels of phosphate and calcium, our study found that all patients with hypocalcemia (100%) had poorly-controlled diabetes, in addition to 84.6% of patients with hypophosphatemia showed poorly-controlled diabetes. These findings emphasize the importance of vitamin D in controlling diabetes. Figure 2 further illustrates this point, showing a significant difference in the average level of vitamin D between the controlled and poorly controlled groups, in which patients with poorly-controlled diabetes had lower levels of average vitamin D as compared to controlled diabetic patients.

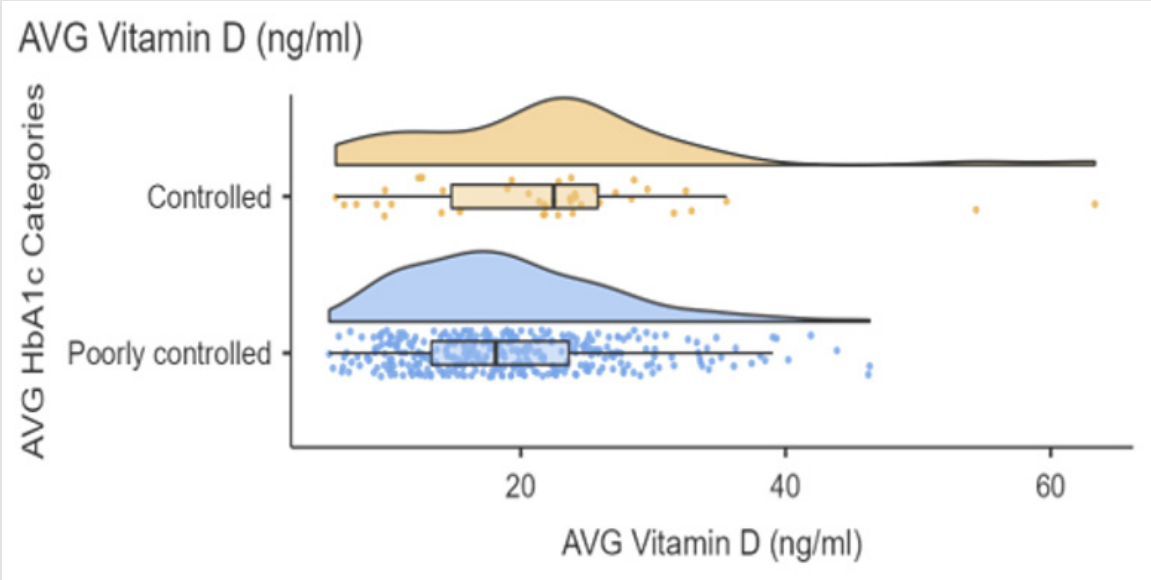

Figure 2: A plot demonstrating the variation and distribution of average vitamin D levels among controlled and poorly controlled diabetic patients.

Table 2: The association between average HbA1c, average vitamin D, phosphate, and calcium levels, divided into categories.

AVG HbA1c Categories		AVG Vitamin D Categories			Phosphate levels Categories				Calcium levels categories			
		Deficiency	Insufficiency	Total	Elevated	Decreased	Normal	Total	Elevated	Decreased	Normal	Total
Controlled	N[a]	15	28	43	6	2	35	43	2	0	41	43
	%	6.6 %	17.0 %	11.0 %	18.8 %	15.4 %	10.1 %	11.0 %	14.3 %	0.0 %	12.0 %	11.0 %
Poorly controlled	N[a]	212	137	349	26	11	312	349	12	35	302	349
	%	93.4 %	83.0 %	89.0 %	81.3 %	84.6 %	89.9 %	89.0 %	85.7 %	100.0 %	88.0 %	89.0 %
Total	N[a]	227	165	392	32	13	347	392	14	35	343	392
	%	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %	100.0 %
X^2		10.5			2.52				4.81			
P-Value		0.001			0.284				0.09			

The Distribution of Vitamin D Levels Among Diabetic Groups

The patients were categorized according to their average HbA1c readings into two groups – controlled and poorly controlled, and as the data is not-normally distributed, the authors used Kruskal-Wallis test to compare both groups in regards of age, gender, BMI, average FBG, average vitamin D, calcium, and phosphate serum levels (Table 3). The study results indicated noteworthy differences in the average levels of vitamin D, phosphate, calcium, and FBG across the two groups. The analysis revealed that FBG levels were notably different among the two comparison groups (P <0.001). Furthermore, the analysis found that the controlled and poorly-controlled groups had significant difference in the levels of vitamin D (P = 0.036). The study conclusively demonstrates that vitamin D deficiency has varying effects on different groups. Moreover, it was found that poorly-controlled diabetes patients showed significantly different serum calcium levels compared to controlled diabetes patients (P=0.008), highlighting the significant impact of vitamin D deficiency.

Table 3: The distribution of average FBS, average vitamin D, calcium, and phosphate levels among controlled and poorly controlled diabetic patients.

	P Value	ε^2
Age	0.114	0.0068
Gender	0.759	2.41E-04
BMI	0.571	8.20E-04
Average FBG (mmol/L)	<.001	0.16827
Average Vit. D (ng/dl)	0.036	0.01128
Calcium level (mmol/L)	0.008	0.01795
Phosphate level (mmol/L)	0.373	0.00203

The Correlation Between Vitamin D and Glycemic Control

To investigate the relationship between vitamin D levels, HbA1c, and FBG, the authors determined the correlation between the mean differences and the correlation between these differences and serum calcium and phosphate levels, using Spearman correlation. Table 4 shows the findings, which reveal a negative correlation (r=-0.27, P <0.001) between the mean difference of vitamin D and the mean difference of HbA1c. Essentially, this indicates that as serum vitamin D levels increase over the course of nine months, HbA1c levels decrease, resulting in better diabetic control. In addition, Figure 3A depicts that patients with a negative vitamin D mean difference (i.e., a decline in serum vitamin D readings over time) will have a positive HbA1c mean difference, indicating that as vitamin D levels decrease, HbA1c levels increase. Figure 3B demonstrates the negative correlation (r=-0.126, P= 0.012) between the mean difference in FBG and vitamin D levels. As shown, the higher the levels of vitamin D, the lesser is the level of FBG.

Table 4: The correlation between vitamin D, HbA1c, FBG mean differences and calcium and phosphate levels.

		HbA1c Mean Difference	FBG Mean Difference
Vitamin D Mean Difference	r	-0.27	-0.126
	P-Value	< 0.001	0.012
HbA1c Mean Difference	r	-	0.304
	P-Value	-	<.001
Calcium Level (mmol/L)	r	-0.81	-0.041
	P-Value	0.11	0.424
Phosphate Level (mmol/L)	r	0.48	-0.034
	P-Value	0.342	0.505

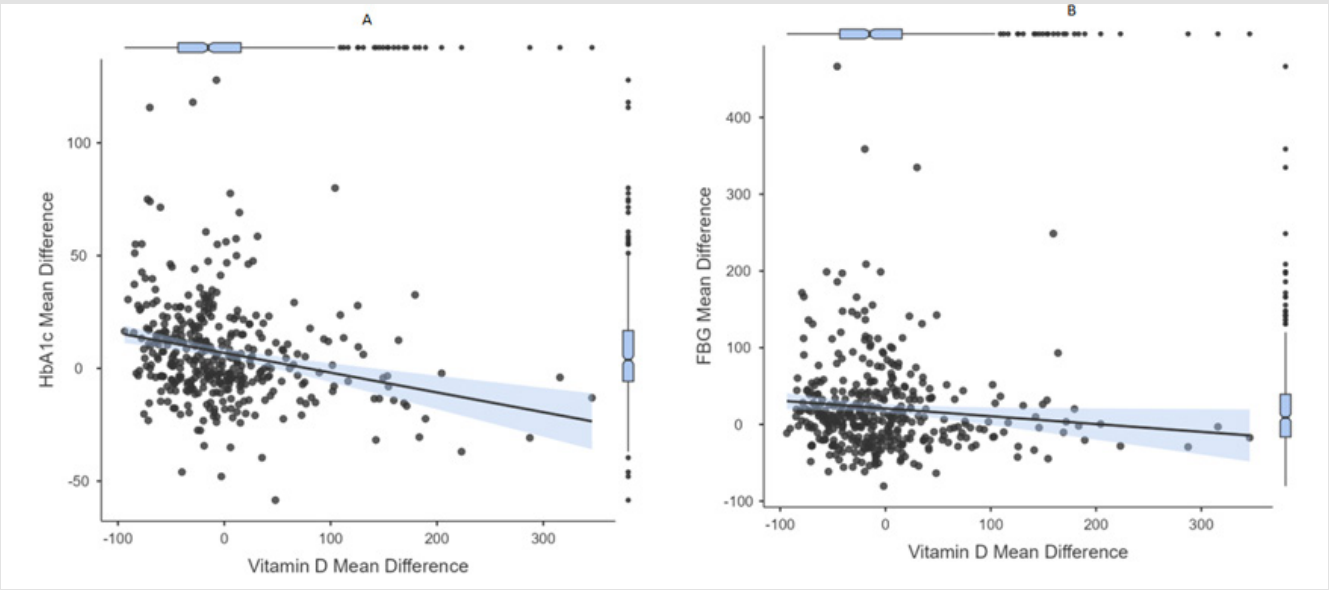

Figure 3:

A. A correlation matrix plot demonstrating the correlation between vitamin D and HbA1c mean difference.

B. A correlation matrix plot between the mean difference of fasting blood glucose (FBG) and vitamin D.

The Predictability between Vitamin D, HbA1c, and FBG

To study whether the mean difference of vitamin D levels can anticipate fluctuations in the mean difference of HbA1c and FBG levels, as well as serum calcium and phosphate levels, a simple linear regression analysis was performed. We considered kidney disease status, patient age, and BMI categories, as factors may influence the findings (Table 5). According to the results of linear regression analysis, the mean difference of vitamin D can significantly predict the mean difference of both fasting blood glucose (FBG) (P=0.033) and glycosylated hemoglobin (HbA1c) (P<0.001). This finding suggests that patients who had increasing levels of vitamin D over the course of 9 months, indicating a positive mean difference, are expected to experience decreasing levels of both HbA1c and FBG, resulting in a positive mean difference.

Table 5: The results of predictability studies between the mean difference of vitamin D, HbA1c, and FBG.

	Model Coefficients - FBG Mean Difference		Model Coefficients – HbA1c Mean Difference	
Predictor	**Beta**	**p**	**Beta**	**p**
Vitamin D Mean Difference	-0.106	0.033	-0.0896	< .001
Calcium levels (mmol/L)	3.3938	0.192	-1.523	0.229
Phosphate levels (mmol/L)	-9.809	0.452	3.8892	0.415
Kidney disease:				
1 – 0	37.893	0.041	0.4576	0.946
Age Categories:				
<50 years – >50 years old	6.737	0.387	-3.6253	0.204
BMI Categories:				
Overweight – Obesity Class I	-7.54	0.338	-4.8711	0.091
Normal Weight – Obesity Class I	7.28	0.593	4.084	0.414
Obesity Class III – Obesity Class I	2.717	0.79	0.2138	0.954
Obesity Class II – Obesity Class I	-6.019	0.478	0.7459	0.81
Obesity Class I – Obesity Class I	26.909	0.443	1.8819	0.933

Discussion

Between March 2023 and October 2023, a total of 392 patients were conveniently collected from the Diabetes and Endocrine center at Prince Hamza Hospital in Amman, Jordan. The data collection process involved obtaining three readings of FBG, HbA1c, Vitamin D, and blood pressure at three months interval, as well as serum levels of calcium and phosphate. This retrospective study aims to investigate the possible association of Vitamin D deficiency and glycemic control status in type II diabetic patients and determine whether it significantly affects glycemic control in these patients. Maintaining proper glycemic control is a crucial aspect of managing diabetes, as it has a significant impact on long-term outcomes and mortality rates and can also help predict a patient›s prognosis. Numerous studies have emphasized the importance of glycemic control in reducing the risk of diabetes-related complications. Basically, the key feature of uncontrolled diabetes-related microvascular disease is angiogenesis, leading to diabetic nephropathy, retinopathy, and neuropathy [12]. The Diabetes Control and Complications Trial (DCCT) and the United Kingdom Prospective Diabetes Study (UKPDS) have demonstrated the benefits of intensive glycemic control on microvascular complications in type 1 and type 2 diabetes, respectively [13,14].

Numerous studies have been conducted to determine the primary factors that can affect glucose levels in individuals with diabetes, which can directly impact the management of the disease. Recent evidence suggests that vitamin D plays a crucial role in glycemic control, including its association with insulin resistance and its potential to interfere with the development of type 2 diabetes mellitus [15-20]. Additionally, vitamin D has been found to affect skeletal muscle metabolism, insulin sensitivity, and lipid composition [21]. Moreover, some evidence suggests that vitamin D levels could affect insulin secretion as well. In a cross-sectional study conducted in Kenya found a significant inverse correlation between vitamin D and glycemic control, as well as vitamin D and BMI among patients with type 2 diabetes, suggesting a potential association between vitamin D deficiency and diabetes control [21]. Furthermore, a review study found that results from recent trials are consistent with a large body of evidence from observational studies indicating that vitamin D plays a role in modulating diabetes risk [22]. This study showed a significant difference in the level of vitamin D between patients with good glycemic control and those with poor glycemic control. Additionally, it is widely recognized in the literature that low serum calcium levels could lead to reduced insulin levels, thus affecting serum glucose levels and glycemic control [22]. Our findings align with these findings, as we observed that uncontrolled diabetic patients had lower levels of serum calcium, which could be attributed to either the low levels of vitamin D or, more importantly, poor glycemic control and reduced insulin levels.

We hypothesized that calculating the mean difference of each variable over the nine-month period would be the most efficient way to explore the correlation between vitamin D, HbA1c, and FBG. Upon conducting this analysis, we discovered a significant inverse correlation between the mean difference of vitamin D and both FBG and HbA1c. Such finding indicates that increasing vitamin D levels over the nine months significantly decreased both FBG and HbA1c, with HbA1c being more affected. This reveals that FBG and HbA1c levels dramatically dropped throughout the nine months as vitamin D levels increased, suggesting that higher vitamin D levels may help with better glycemic control. Our findings align with those of Lips P, et al. [21] who also found a negative correlation between vitamin D levels and diabetic control status [23]. Neck et. Al (2023) suggested that poorly controlled diabetic patients had significantly lower mean vitamin D levels compared to those with good glycemic control, indicating a potential association between vitamin D and glycemic control [24]. According to our research, a decrease in vitamin D levels can be a significant predictor of poor glycemic control, as indicated by a positive mean difference in Hb1Ac. Conversely, an increase in vitamin D levels can predict better glycemic control ($P < 0.001$). Additionally, we observed a statistically significant linear correlation between vitamin D levels and FBG ($P=0.033$), implying that vitamin D levels could predict FBG levels.

The correlation between vitamin D levels and glycemic control remains a topic of debate and investigation [25]. The findings of previous literature are inconsistent, as certain studies have indicated a significant association between the two, others have shown no observable correlation [25,26]. This implies that the impact of vitamin D supplementation on glycemic control may not be noticeable in every diabetic patient and could be influenced by covariates such as diabetes type and personal characteristics [26]. Additionally, vitamin D deficiency is highly prevalent among individuals, which could be seen as a part of metabolic panel among diabetic patients, regardless of their glycemic control [24,27,28]. This is the first study in Jordan that investigated the possible association between vitamin D and glycemic control in diabetic patients. However, our study faced some limitations; we collected the patients conveniently, and followed them retrospectively for a limited duration of time, in addition to the possible various confounding bias. Thus, additional comprehensive clinical trials and long-term follow up studies of diabetic patients while monitoring their vitamin D status are necessary to elucidate the specific role and the therapeutic possibilities of vitamin D in enhancing glycemic control in diabetic individuals.

Conclusion

Vitamin D deficiency is associated with poor glycemic control among Jordanian patients, and the levels of serum vitamin D could predict the glycemic status in these patients. However, the association could be affected by many confounding factors, and the need for further precise studies to elaborate on the association is warranted.

Highlights

- Vitamin D deficiency and type II diabetes.
- Association between vitamin D and glycemic control.
- The effect of vitamin D levels on serum fasting blood glucose and HbA1c
- Retrospective study in Jordan.
- The predictability between vitamin D levels and fasting blood glucose and HbA1c.

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