

Status of Vitamin D in Individuals with Type 2 Diabetes Mellitus

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ABSTRACT

Background: Recently, there has been an increasing interest towards the metabolic and clinical effects of vitamin D deficiency. Vitamin D has been suggested to impact glucose homeostasis and, thus, metabolic syndrome and type 2 diabetes mellitus (T2DM) seem to be linked to vitamin D deficiency.

Aim: To study serum "25-hydroxy vitamin D" levels in a group of T2DM patients and to investigate its possible influence on the degree of glycemic control and lipid metabolism.

Methods: This is a case- control study which enrolled 65 known cases of T2DM and 65 non- diabetic subjects. Blood levels of vitamin D, glycosylated hemoglobin (HbA1c), fasting serum lipids and plasma glucose (FPG) were measured. The impact of vitamin D levels on the extent of glycemic control and serum lipids was investigated.

Results: The mean serum level of vitamin D was significantly lower among T2DM patients compared to controls ($p < 0.0001$). Vitamin D was significantly and inversely correlated with HbA1c and FPG values in diabetic patients ($r = -0.54$ and -0.578 respectively, $p < 0.0001$). In addition, vitamin D showed negative correlations with serum triglycerides, total cholesterol, VLDL- cholesterol and low density lipoprotein cholesterol (LDL- c), but significant positive correlation with high density lipoprotein cholesterol (HDL- c).

Conclusions: Patients with T2DM frequently have vitamin D deficiency. Vitamin D insufficiency may be linked to poor glycemic control and dyslipidemia in T2DM patients. Vitamin D therapy may, thus, improve glycemic control in type 2 diabetes mellitus and ameliorate metabolic derangements.

Keywords: vitamin D, HbA1c, T2DM, lipids, FPG.

حالة فيتامين (د) لدى الأفراد المصابين بمرض السكري من النوع الثاني

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الخلاصة

الخلفية: في الآونة الأخيرة، كان هناك اهتمام متزايد نحو الآثار الأيضية والسريرية لنقص فيتامين (د). هناك اعتقاد أن فيتامين د يؤثر على توازن الجلوكوز، وبالتالي، يبدو أن متلازمة التمثيل الغذائي ومرض السكري من النوع ٢ مرتبطان بنقص فيتامين د.

الهدف: هو دراسة مستويات "٢٥-هيدروكسي فيتامين د" في الدم لدى مجموعة من مرضى السكري من النوع ٢ ودراسة تأثيره المحتمل على درجة التحكم في نسبة السكر في الدم واستقلاب الدهون.

الطرق: هذه دراسة الحالات والشواهد التي سجلت ٦٥ حالة معروفة من مرضى السكري من النوع ٢ و ٦٥ شخصا غير مصابين بالسكري. تم قياس مستويات فيتامين د والهيموجلوبين الغليكوزيلاتي (HbA1c) والدهون في مصل الدم الصائم وجلوكوز البلازما (FPG) في الدم. تمت دراسة تأثير مستويات فيتامين د على مدى التحكم في نسبة السكر في الدم والدهون في الدم.

النتائج: كان متوسط مستوى فيتامين د في المصل أقل بشكل ملحوظ بين مرضى السكري من النوع ٢ مقارنة بالمجموعة الضابطة ($P > 0.0001$). ارتبط فيتامين د بشكل ملحوظ وعكسي مع قيم الهيموجلوبين الغليكوزيلاتي وجلوكوز البلازما لدى مرضى السكري ($r = -0.54$ و -0.578 على التوالي، $p < 0.0001$). بالإضافة إلى ذلك، أظهر فيتامين (د) ارتباطات سلبية مع الدهون الثلاثية في الدم، والكوليسترول الكلي، وال كوليسترول البروتين الدهني منخفض الكثافة جدا VLDL- وكوليسترول البروتين الدهني منخفض الكثافة (LDL-c)، ولكن ارتباطاً إيجابياً كبيراً مع كوليسترول البروتين الدهني عالي الكثافة (HDL-c).

الاستنتاجات: المرضى الذين يعانون من مرض السكري من النوع ٢ كثيرا ما يعانون من نقص فيتامين (د). قد يرتبط نقص فيتامين د بضعف التحكم في نسبة السكر في الدم واضطراب شحوم الدم لدى مرضى السكري من النوع ٢. وبالتالي فإن العلاج بفيتامين د قد يحسن التحكم في نسبة السكر في الدم لدى مرضى السكري من النوع ٢ ويخفف من الاضطرابات الأيضية.

الكلمات المفتاحية: فيتامين د، الهيموجلوبين السكري التراكمي، مرض السكري النوع الثاني، الدهون، سكر الدم الصائمي.

INTRODUCTION

The sunshine vitamin- vitamin D- plays a crucial role in the creation of healthy well-calcified vertebrates' skeletons as well as having a control over the production and the action of some hormones (like estrogen and progesterone). When exposed to sunshine, 7-dehydrocholesterol in the skin is transformed into previtamin D3, which then quickly isomerizes to vitamin D3. Once produced, vitamin D3 is converted to 25-hydroxyvitamin D3 (the main vitamin D reservoir) in the liver, and to 1, 25-dihydroxyvitamin D3 in the kidneys, which is its active form and is also known as calcitriol. Calcitriol, which is considered as a hormone, plays an important role in the metabolism of calcium and phosphorus and maintain bone health¹.

Vitamin D interacts with its wide-spread receptors (VDR) to elucidate its biological effects. Many diseases have been linked to insufficient vitamin D levels and/or defective D/ VDR interactions. Maintaining appropriate levels of Vitamin D can be a helpful preventive strategy pathophysiologically. Early-life vitamin D supplementation has been shown to lower risk of several diseases, with immune-protective effects². Diabetes mellitus (DM) is a metabolic condition characterized by chronically high blood glucose levels³. Around the world, the incidence of diabetes is expected to rise by about ten percent by 2030⁴, and is considered as a big health concern. By now, DM is considered as the tenth most common cause of death worldwide, causing around one million fatalities every year⁵. Good glycemic control in diabetes can lower risk of macrovascular and microvascular complications. In fact, HbA1c is a universally accepted measure for long-term glycemic control monitoring that not only indicates the likelihood of complications development, but also serves as a standalone heart disease risk factor^{6,7}.

Over eighty- seven percent of handicapping in low- income developing nations can be attributed to type 2 diabetes mellitus (T2DM), which is also a known cause for dyslipidemia and hypertension^{8,9}. In addition, it had been shown that pre-diabetes is linked to a higher risk for various heart diseases¹⁰.

People with T2DM frequently have abnormal lipid profile which is referred to "diabetic dyslipidemia" and the frequencies of various lipids abnormalities can vary depending on ethnicity, socioeconomic status, and access to medical treatment¹¹⁻¹⁴.

According to a recently released meta-analysis, aberrant levels of serum lipid could partially predict risk of diabetes¹⁵.

Investigations of individuals with T2DM revealed significant correlations between atherosclerosis and high serum triglycerides as well as low high density lipoprotein- cholesterol (HDL-c) levels together^{16,17}. The aim of this work is to study serum vitamin D levels among Iraqi diabetic patients and to investigate its possible relations with serum lipids and the extent of glycemic control as revealed by blood fasting plasma glucose (FPG) and HbA1c levels.

MATERIALS AND METHODS

This is a case- control study that was approved by the Medical Research Ethical Committee, College of Medicine, University of Mosul (UOM/COM/MREC/22-23/JL6). All participants were introduced to the study objectives and procedures prior to their consent to participate and the study was conducted in accordance to declaration of Helsinki regarding the ethical principles for medical research in humans.

A total of 130 subjects participated in this study. They aged 25 to 71 years and were divided into two groups; apparently healthy non- diabetic controls (n=65) and known cases of T2DM on regular hypoglycemic medications (n=65 people). Cases were collected from private clinics. The inclusion criteria for the group of diabetic cases were:

1. Known cases of T2DM of any duration
2. Age of 25 yr and above
3. Consent to participate (verbal or written)

However, cases of T2DM were excluded upon having one or more of the followings:

- Type 1 diabetes mellitus
- 1. Receiving vitamin D supplements
- 2. Underlying liver and kidney diseases.
- 3. Malignant conditions and those with thyroid or parathyroid abnormalities
- 4. Taking any medications that interacts with vitamin D or affects its metabolism.

The control group enrolled apparently healthy age and sex- matched subjects without any history of type 1 or type 2 DM. They had no any history of liver, kidney, thyroid and parathyroid diseases.

They were not receiving any vitamin D supplements or any medicines that interfere with vitamin D metabolism and consented to volunteer.

All subjects were interviewed and asked to fast overnight prior to blood collection as appointed. A 7- ml venous whole blood sample was collected from each participant where two ml were transferred into EDTA containing tubes for HbA1c estimation, one ml was placed into sodium fluoride/potassium oxalate tubes for plasma glucose assessment while the rest 4 ml were transferred into plain tubes, allowed to clot and centrifuged at 4000 rpm for 5 minutes for serum separation. Serum was aliquoted and frozen at – 20 °C for subsequent analysis of fasting lipid profile and vitamin D determination.

Serum lipids, FPG and HbA1c levels were measured with the principle of electrochemiluminescence using Cobas c 111 automated clinical chemistry analyzer and reagents (Roche Diagnostics), in accordance with manufacturer's instructions. However, serum levels of 25-hydroxyvitamin D were determined using Cobas e 411 chemistry analyzer. According to the guidelines of the Endocrine Society and the Institute of Medicine, 25-hydroxyvitamin D levels below 20 ng/mL indicated a deficiency, 20- 30 ng/mL indicated insufficiency, and over 30 ng/mL were considered normal 18.

Statistical Analysis

Data were analyzed using the Statistical Package of Social Sciences (SPSS) program (version 20.0). Descriptive statistics were used to obtain the mean, standard deviation (SD) and range (minimum- maximum) values of the studied variables. Unpaired Student's t-test was employed to compare means of continuous parameters between cases and controls. Chi- square test was, however, used to compare categorical (non-continuous) variables as indicated. Pearson correlation test was performed to indicate whether vitamin D levels are correlated to FPG, serum lipids and HbA1c or not. Statistical significance was considered at $p \leq 0.05$.

RESULTS

Basic Characteristics of the Study Subjects

One hundred- thirty volunteers in total participated. They formed two groups; 65 T2DM patients (cases) and 65 non-diabetics (controls). The T2DM cases were 30 females and 35 males with mean age of 53.45 ± 10.48 years. However the control group was comprised of 36 females and 29 males with the mean age of 45.66 ± 12.17 years, Table (1).

Table (1): Age and sex distribution of T2DM cases and non- diabetic controls

		T2DM (N=65)	Controls (N=65)	p-value
Age (Years)	Mean± SD	53.45 ± 10.48	45.66 ± 12.17	0.0001
	Range (min- max)	34- 71	30-65	
Sex	Females	No.	30	36
		%	46.15	55.38
	Males	No.	35	29
		%	53.85	44.61

Comparisons of Biochemical Variables

When the mean values of the measured biochemical variables of diabetics were compared to those corresponding values of controls, it was revealed that FPG, serum total cholesterol, LDL-c, VLDL-c and triglycerides mean values in addition to HbA1c were significantly higher in the T2DM patients' group. However, HDL-c values were significantly lower among cases compared to control. The mean serum vitamin D level among diabetics was significantly lower than that of the control group (15.11 ± 6.71 Vs 29.79 ± 12.34 ng/mL, $p < 0.0001$), Table (2).

Table (2). Comparison of serum vitamin D, lipid profile, HbA1c and FPG mean values of the study groups. Data are expressed as mean± SD.

	T2DM (N= 65)	Controls (N= 65)	p-value
Vitamin D (ng/mL)	15.11 ± 6.71	29.79 ± 12.34	<0.0001
FPG (mg/dL)	222.25 ± 94.96	97.74 ± 9.97	<0.0001
HbA1c (%)	8.55 ± 2.0	5.57 ± 0.33	<0.0001
Total cholesterol (mg/dL)	206.35 ± 54.27	164.86 ± 54.27	<0.0001
HDL-c (mg/dL)	43.11 ± 4.98	48.19 ± 4.20	<0.0001
LDL-c (mg/dL)	120.65 ± 47.85	91.09 ± 29.0	<0.0001
VLDL-c (mg/dL)	46.17 ± 36.0	25.16 ± 9.10	<0.0001
Triglycerides (mg/dL)	211.23 ± 115.76	119.34 ± 35.71	<0.0001
Atherogenic index	5.51 ± 4.83	3.38 ± 0.68	<0.001

Stratification of T2DM Patients and Controls by Vitamin D State

In referral to the Endocrine Society and the Institute of Medicine guidelines, as was already described, about 71% of DM patients got vitamin D deficiency, 29% showed insufficiency and 0% got adequate vitamin D. Table (3) compares the distribution of vitamin D state in the studied diabetic patients and controls.

Table (3). Stratification of T2DM cases and controls by serum vitamin D levels. Data are expressed as N (%).

	T2DM (N=65)	Controls (N=65)	p-value
Deficiency (Vitamin D less than 20 ng/mL)	46 (70.76)	17 (26.15)	<0.00001
Insufficiency (Vitamin D 20- 30ng/mL)	19 (29.23)	13 (20)	
Adequate (Vitamin D > 30 ng/mL)	0 (0)	35 (53.84)	

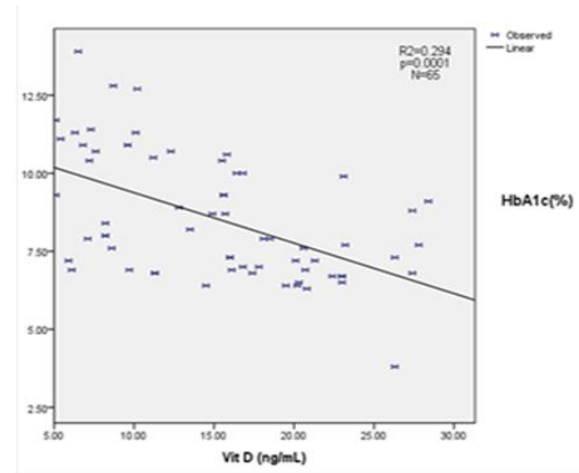
Relation of Serum Vitamin D with Indicators of Glycemic Control

Among the diabetic patients, serum vitamin D levels were negatively and significantly correlated with levels of FPG and HbA1c (as short and long-term glycemic control indicators) respectively, Table (4) and Figure (1).

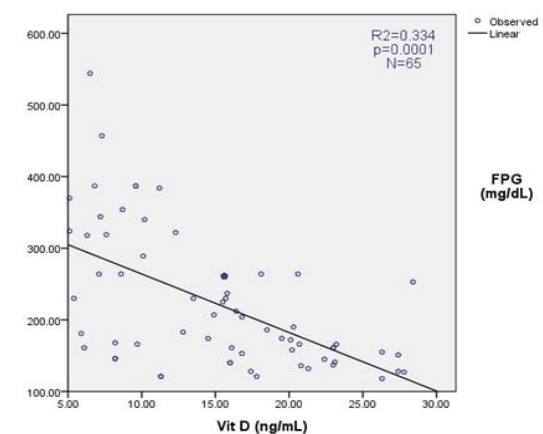
Table (4). Correlation between serum vitamin D levels and glycemic control indicators among cases and controls.

	T2DM (N=65)		Controls (N=65)	
	r	p-value	r	p-value
HbA1c (%)	-0.542	0.0001	-0.047	0.71
FPG (mg/dL)	-0.578	0.0001	-0.069	0.05

*Using Pearson correlation analysis.



A



B

Figure (1). Linear regression analysis of serum vitamin D with HbA1c levels (A) and FPG levels (B) among T2DM patients.

Relation of Serum Vitamin D with serum lipids

Using Pearson's correlation test, serum vitamin D levels were negatively and significantly correlated with serum fasting levels of total cholesterol, LDL-c, VLDL-c and triglycerides among diabetic patients. However, HDL-c was significantly and positively correlated with serum vitamin D levels, Table (5) and Figures (2-4).

Table (5). Correlation of serum vitamin D levels with serum lipid profile among diabetic patients.

	T2DM (N=65)	
	r	p-value
Total cholesterol	-0.123	0.328
LDL-c	-0.067	0.594
VLDL-c	-0.326	0.008
HDL-c	0.301	0.015
Triglycerides	-0.387	0.000

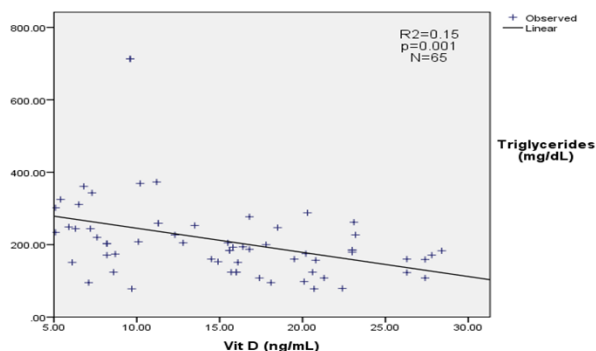


Figure (2). Linear regression analysis of serum vitamin D and triglycerides levels among patients with T2DM

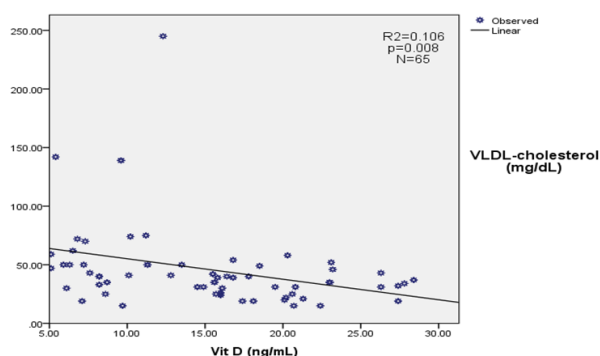


Figure (3). Linear regression analysis of serum vitamin D and VLDL-cholesterol levels among patients with T2DM

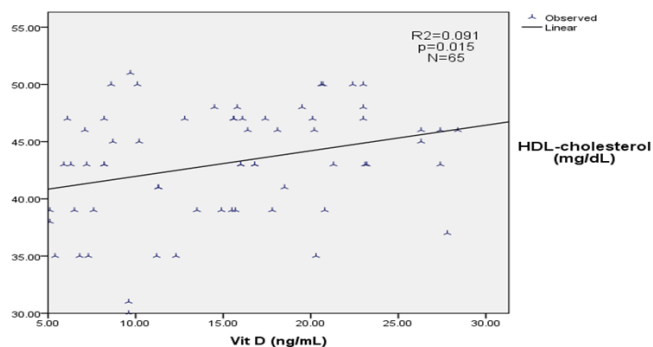


Figure (4). Linear regression analysis of serum vitamin D and HDL-cholesterol levels among patients with T2DM

DISCUSSION

The connection between vitamin D and different metabolic disorders including T2DM has been the subject of concern worldwide recently. Vitamin D is linked to the breakdown of glucose, the emergence of type 2 diabetes, and the so-called metabolic syndrome¹⁹⁻²¹. A research has suggested that insufficient vitamin D intake may increase the likelihood of developing T2DM and metabolic syndrome²².

In the current study, it was revealed that diabetic patients had significantly lower mean serum level of "25- hydroxyvitamin D" compared to non-diabetic controls (Table 2). Researchers had revealed that lack of vitamin D resulted in reduced insulin release and that its re-supply enhanced pancreatic β -cell activity and restored glucose tolerance²³. Chiu *et al.*, in agreement with our study, had also concluded that lack of vitamin D increased the chance of developing diabetes and metabolic syndrome²⁴.

A rising number of studies point to a connection between vitamin D state and risk of type 2 diabetes. Palomer *et al.*

showed that having T2DM may be more likely among those with insulin resistance and low vitamin D levels²⁵.

The current study showed significantly higher levels of some serum lipids (total cholesterol, LDL-c, VLDL-c and triglycerides) among diabetic patients compared to controls. This is attributed to resistance to insulin and its shortage which influence the important enzymes and molecular pathways of lipid metabolism. Such alterations in blood lipids in diabetes were also the findings of other investigators²⁶. Increased LDL-c, reduced HDL-cholesterol and/ or higher triglycerides levels are all examples of dyslipidemia in diabetes mellitus²⁷.

The two disorders, dyslipidemia and vitamin D insufficiency, are extremely common and independently linked to several cardiovascular risk factors²⁸.

An observational study has shown that adequate levels of vitamin D are linked to a healthy lipid profile, while low vitamin D levels are linked to an atherogenic lipid profile²⁹.

According to the current study, vitamin D deficiency was considerably more frequent in type 2 diabetic patients than in the control group (70.76% Vs 26.15%), while insufficiency affected 29.23% of patients compared to 20% of controls, Table (3). This is in agreement with Khudayar *et al.* who demonstrated that vitamin D deficiency is more common in people with type 2 DM, and that vitamin D supplementation may result in a better glycemic control³⁰.

Lips *et al.*, in their review, also showed that low vitamin D is associated with diabetes. They explained this association by two concepts; first, vitamin D stimulates insulin secretion from pancreatic B cells (thus, vitamin D deficiency is associated with insulin resistance), and second, vitamin D deficiency causes inflammation and increases inflammatory markers.

Moreover, it has been found that genetic polymorphism of VDRs may lead to impaired glycemic control³¹.

Intriguingly, an inverse relationship between vitamin D levels and glycemic control estimates (HbA1c and FPG) was pointed out in our study (Table 4, Figure 1; A&B). It seems that vitamin D may be involved in glucose homeostasis in T2DM. This is consistent with the outcomes of Laway *et al.* who noticed a significant negative relationship between vitamin D and HbA1c among diabetics³². According to the study of Lau *et al.*, there was also a negative association between vitamin D levels and glycosylated hemoglobin in women with gestational diabetes³³.

In their study, Pittas *et al.*, found that supplementing non-diabetic elderly people with 700 IU of vitamin D plus 500 mg of calcium daily over three years resulted in a significantly lower extent of elevation in plasma glucose levels than in those received a placebo. This finding further supports the link between vitamin D deficiency and diabetes' likelihood³⁴.

In this study, there were negative correlations between vitamin D levels and total cholesterol, LDL-c, VLDL-c and triglyceride among cases of T2DM (statistically significant regarding VLDL-c and triglycerides), Table 5 and Figures (3 &4). This is in agreement with Elshebiny *et al.*³⁵.

On the other hand, a significant positive correlation was seen between vitamin D and HDL-cholesterol levels in the cases group (Table 5 and Figure 4). This is, again, in agreement with Elshebiny *et al.* and Saedisomeolia *et al.*, who also revealed that serum levels of vitamin D had a positive correlation with HDL-cholesterol^{35,36}.

CONCLUSIONS

Our research clearly shows an unfavorable relationship between vitamin D levels and state of glycemic control in diabetes as reflected by HbA1c and plasma glucose. People with Type 2 diabetes with inadequate vitamin D had a higher chance of developing dyslipidemia. Therefore, vitamin D testing should be recommended mainly for those diabetics with poor glycemic control. This may further support the need for vitamin D supplementation and increased sun exposure for a better glycemic control and lower rate of metabolic derangements.

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Funding

None.

Conflict of Interest

None

Ethical Consideration

University of Mosul, College of Medicine, Medical Research Ethical Committee approved the project (UOM/COM/MREC/22-23/JL6).

Author's Contribution

Role	Contributor 1	Contributor 2	Contributor 3	Contributor 4
Concepts	√	√	√	√
Design	√	√	√	√
Definition of intellectual content	√		√	
Investigation	√			√
manuscript writing,	√			
Literature search	√	√	√	
Clinical studies	√			√
Data analysis	√		√	
Statistical analysis	√		√	
Manuscript editing	√		√	
Manuscript review	√	√	√	√

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