Estimation of vitamin E level and its relation to lipid profile in patients with type II Diabetes Mellitus

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ABSTRACT

Background: Type 2 diabetes mellitus (T2DM) is considered a global disease as it affects over 150 million people worldwide, a number that is supposed to be doubled by 2025. High glucose levels, in vitro, appear to raise the extent of LDL oxidation, and glycated LDL is more prone to oxidative modification.

Objective: To investigate the relationship between serum level of vitamin E and lipid profile in patients with type II DM.

Methods: This study involved 28 patients suffering from type II DM diagnosed 1-4 years ago and with age ranged from 17 -60 years old, with different residence around Basra ; In addition to 56 apparently healthy persons matched in age and sex to the patients as a control group. The medical histories were taken and General examinations were done to them with measurement of their height and weight. Four milliliters of venous blood was drawn from each patient and control to measure TG, TC, VLDL, HDL, LDL, HbA1C and Vitamin E and the results were used to assess the presence of any association between Vit E levels and lipid profile.

Results: The mean levels for patients and controls of TG were (157.1± 24.9 and 119.8 ± 29.0) mg/dl, of TC were (204.0 ± 43.2 and 168.0 ± 31.5) mg/dl, of VLDL was (31.5 ± 5.0 and 24.2 ± 5.9) mg/dl, of LDL was (129.6 ± 42.8 and 97.6 ± 30.8) mg/dl, of HDL was (42.9 ± 10.2 ± 46.0 ± 9.0) Mg /dl, of HbA_{1C} were (9.4 ± 3.0 and 5.0 ± 0.7) % and of vitamin E were (3.7 ± 1.3 and 6.8 ± 2.0) mg/dl, respectively. There was a significant difference between all the above measured parameters, except of HDL, between cases and controls. Also there was a significant association between DM type II and low level of vitamin E. there was a significant negative correlations between vitamin E and TG, TC, LDL, VLDL but not HDL, even after adjustment for age, sex and BMI.

Conclusion: Diabetes mellitus type II has a significant effect on vitamin E which is an important guard against dyslipidemias, one the major causes of diabetes vascular complications.

Keywords: DM type II, Vitamin E, Dyslipidemia, diabetes mellitus.

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Type 2 diabetes mellitus (T2DM) is considered a global disease as it affects over 150 million people worldwide, a number that is supposed to be doubled by 2025 [1].

Development of type II DM is known to be associated with increase in the oxidative stress during its early stages[2]. Also oxidative stress is blamed to be involved in the development of diabetic complications, mainly the cardiovascular ones [3, 4]. High glucose levels, in vitro, appear to raise the extent of LDL oxidation, and glycated LDL is more prone to oxidative modification [5].

The harmful effect of increasing glucose levels, that is may occur through its effect on fatty acids, is thought to be mediated to largely via increased production of reactive oxygen species (ROS) and reactive nitrogen species (RNS) resulting in increased oxidative stress [3].

Beta Cells are especially susceptible to ROS, as they contain levels of free-radical quenching (antioxidant) enzymes such as superoxide dismutase, glutathione peroxidase, and catalase [6, 7].

On the other hand, ROS also impair delivery of glucose to peripheral tissues, and enhance systemic inflammation [4]; both of these effects are considered an important in future complications.

Vitamin E (Tocopherol) is the most important fat soluble vitamin with antioxidant and anti-inflammatory activities, and if it is given at the appropriate dose and form, it would have beneficial effects on cardiovascular disease in a high-risk population such as diabetic patients [8], also In T2DM patients, vitamin E was found to reduce the serum levels of IL-1β, IL-6, TNF-α, PAI-1, and CRP [9] exerting an increasing benefit in these patients.

Diabetes Mellitus is commonly associated with dyslipidemia, because both insulin deficiency and resistance affect enzymes and pathways of lipid metabolism [10]. The dyslipidemia of type 2 DM is characterized by high triglyceride (TG) levels, low athero-protective high density lipoprotein-cholesterol (HDL-C) levels, and high levels of small dense low density lipoprotein-cholesterol (LDL-C) [11, 12].

LDL is a major vehicle for both cholesterol and vitamin E delivery to peripheral tissues; this facilitates its role as a powerful antioxidant guard against "oxidized LDL", which is thought to be a significant precursor to atherosclerosis[13].

Methods. Eighty four total subjects were included in this case control study which was carried out from the first of November 2013 to the end of January 2014: 28 patients with type two DM diagnosed 1- 4 years ago with different

levels of glycemic control (HBA_{1C} ranged from 5.5 – 15.5 %), their age ranged from 17-60 years, from different residence around Basra; in addition to 56 apparently healthy controls who are matched to the patients in age and sex. Any patient known to have vitamin supplement were excluded. Also we exclude all patients with psychological disorders.

The medical history was taken from the patients, with emphasis on history of hypertension, smoking and any family history of DM type II.

General examinations were done to the patients, and the height and weight of all subjects were measured to calculate the BMI.

Four milliliters of venous blood was drawn from each patient and control to measure cholesterol, TG, HDL-C (bioMerieux sa, France) and calculate VLDL and LDL. Also HBA_{1C} (Human, Germany) and Vitamin E were measured by spectrophotometry.

The results were expressed in form of mean ± standard deviation (SD). The difference between the means of any parameter in study in different groups was assessed by the use of independent sample t-test. The association among categorical variables was assessed by use of chi square test. The correlation between two different parameters was assessed by Pearson's correlation coefficients and Stepwise regression analysis was performed to identify the strongest predictor of DM type II. $P < 0.01$ was considered the lowest limit of significance.

Results. The final study population consisted of 28 patients (male: 12 patients; female: 16 patients) and 56 control (male: 24 control; female: 32 control). General patient characteristics is shown in Table 1

Table 1: General Subjects Characteristics

	Patients (No:28)	Controls (No:56)	P value
Age (years)	45.8 ± 8.8	45.8 ± 10.6	0.994
Sex (male:female)	12:16	24:32	1.000
Weight (kg)	70.4 ±12.6	71.8 ± 14.6	0.684
BMI (kg/m^2)	27.3 ± 5.7	27.6 ± 5.8	0.801
Family history of DM (%)	19 (68%)	31 (55%)	0.271
Mean BP (mmHg)	122 ± 15	115 ± 19	0.127

The mean levels for patients TG, TC, VLDL, LDL, HDL HbA_{1C} and Vitamin E were (157.1 mg/dl, 204.0 mg/dl, 31.5 mg/dl, 129.6 mg/dl, 42.9 mg/dl, 9.4% and 3.7 mg/dl), respectively; while those of the controls were (119.8 mg/dl, 168.0 mg/dl, 24.2 mg/dl, 97.6 mg/dl, 46.0 mg/dl, 5.0 % and 6.8 mg/dl), respectively. All the parameters of lipid profile, apart from HDL, were significantly higher in the patients than the controls. Vitamin E level was significantly lower and HbA_{1C} higher in the diabetic than the control group. Table 2

Regarding effect of sex, there was no difference in level of vitamin E between males and females; but both sexes showed significant difference in vitamin E level between cases and controls. Table 3

Subjects were divided into two group, those with low vitamin E level (< 5.5 mg/dl) and those with normal vitamin E level (≥ 5.5). There was a significant association between low Vit E level and presence of DM type II, as 25 diabetic patients (89%) had low vitamin E level. Table 4

Regarding Pearson's rank correlation test, Vitamin E showed significant negative correlations with HbA_{1C}, TG, TC, VLDL, LDL but not HDL, even after adjustment for age sex and BMI. Also HbA_{1C} showed significant positive correlations with TG, TC, VLDL, and LDL after adjustment for age, sex and BMI.

Table 2: Biochemical Measures.

	Patients (No:28)	Controls (No:56)	P value
Triglyceride (mg/dl)	157.1 ± 24.9	119.8 ± 29.0	< 0.001
Total Cholesterol (mg/dl)	204.0 ± 43.2	168.0 ± 31.5	< 0.001
VLDL Cholesterol (mg/dl)	31.5 ± 5.0	24.2 ± 5.9	< 0.001
LDL Cholesterol (mg/dl)	129.6 ± 42.8	97.6 ± 30.8	< 0.001
HDL Cholesterol (mg/dl)	42.9 ± 10.2	46.0 ± 9.0	0.167
HbA_{1C} (%)	9.4 ± 3.0	5.0 ± 0.7	< 0.001
Vitamin E (mg/dl)	3.7 ± 1.3	6.8 ± 2.0	< 0.001

Table 3: Effect of Sex on Vitamin E levels.

Vitamin E (mg/dl)	Male (No=36)		P value	Female (No=48)		P value
	Patients (No=12)	Controls (No=24)		Patients (No=16)	Control (No=32)	
	3.3 ± 1.2	6.8 ± 1.6	< 0.001	3.9 ± 1.3	6.9 ± 2.4	< 0.001

Table 4: Association between Vitamin E and DM type II.

		Vitamin E level		Total
		Low	Normal	
Subjects	Cases	25	3	28
	controls	15	41	56
Total		40	44	84
χ^2= 29.2		*P* value < 0.001		O.R.= 6 - 86.6

Table 5: Correlation of Vitamin E and HbA1c with Lipid Profile.

Control Variables		HbA_{1C}	TG	TC	VLDL	LDL	HDL
BMI, Age and Sex	HbA_{1C}	1.000	0.427**	0.351**	0.435**	0.293**	-0.31
	Vitamin E	-0.512**	-0.359**	-0.325**	-0.345**	-.0300**	0.086

** Correlation is significant at *P* value < 0.01

Stepwise multiple regression analysis of several factors, including Age, BMI, BP, Vitamin E and different lipoproteins levels, revealed Vitamin E to be the strongest predictor of DM type II (R^2 = 0.402, P < 0.001) followed by TG (R^2=0.508, P < 0.001)

Discussion. T2DM is a multifactorial disease characterized by chronic hyperglycemia, decreased insulin secretion, and diminished responsiveness to normal concentrations of circulating insulin; i.e. insulin resistance [14,15]. Obesity

and the metabolic syndrome are important steps in the evolution of T2DM; and their association with oxidative stress indicates a potential pathogenic role for oxidative agents in the progression of the disease[16].

Various cross-sectional and interventional studies found that consumption of micronutrients has been associated with lower levels of oxidative stress, proinflammatory cytokines, and risk of T2DM [16-18].

This study showed that diabetic patients, who were matched in age, sex and BMI to the control group, had significantly higher TG, TC, VLDL and LDL than the control group. These results are similar to many studies [19-23] which characterize diabetic dyslipidemia.

While few studies found that LDL is decreased only in subgroups diabetic patients[10,24] ; As diabetes is metabolically heterogeneous. Thus the roles of insulin deficiency, insulin resistance, obesity, and genetic factors all may contribute the heterogeneity in lipoproteins levels.

Serum level of HDL was lower in diabetic patients than the control which is similar to that of Gordon et al and Ehimen et al studies[20,21] but the difference didn't reach statistical significance , possibly due to small sample size.

Diabetic dyslipidemia is one of the most important contributors to increase risk of Coronary heart disease in these patients. The etiology of this dyslipidemia is multifactorial; there may be decrease in activity of lipoprotein lipase leading to hypertriglyceridemia and increase in Chylomicron and VLDL remnants, also hyperglycemia may interfere with removal of triglyceride – rich lipoproteins and thus accentuating hypertriglyceridemia [22], on the other hand increase in certain cytokines in diabetic patients (mainly IL-1 and IL-6) stimulate the liver to increase VLDL secretion [19]. Recently, it was found that gyration of several major or minor apolipoproteins, apo E phenotype frequency, free cholesterol or triglyceride enrichment of VLDL and LDL may the bases to structural rather than quantitative alteration in lipid profile [23].

Diabetic patients showed significantly lower serum level of vitamin E than the control with 25 patients (89%) with vitamin E below normal level indicating a significant association with type II DM, these results consistent with those of other studies [2, 20, 25-27].

The decrease in vitamin E is more probably due to depletion during the process of combating the increased free radicals that are generated in diabetes [20].

Vitamin E has a lot of beneficial effect in patients with DM type II; as it was found to reduce the serum levels of IL-1β, IL-6, TNF-α, PAI-1, and CRP, protects LDL from peroxidation, and improve T2DM-associated abnormal metabolic patterns (hyperglycemia, dyslipidemia, and elevated levels of FFAs) [16]

Despite the above mentioned facts, the results of vitamin E supplementation on prevention and treatment of type II DM are controversial; Mark and Yoichi, [28] by systematic review and meta-analysis of prospective cohort studies, reported that intake of antioxidants, mainly vitamin E was associated with a 13% reduction in the risk of type 2 diabetes mellitus , while other studies [29,30] found no such benefit. This may simply result from genetic differences between individuals that could lead to different degree of response to micronutrient exposure [31], or may highlight the need to intervene with most appropriate antioxidant combination and at the most suitable moment to the patient at risk [32].

There was significant correlation between level of vitamin E and HbA1c in this study and that differs from the results of other studies [33, 34] however two studies found that vitamin E supplementation improve HbA1c [18, 35] and evidence suggests that poorly controlled diabetic patients with low serum vitamin E would benefit from vitamin E supplementation [36].

Few studies assess the correlation between tocopherol level and lipid profile. Lopes et al.[37] found that positive correlation between vitamin E and TG.in smokers. Gupta et al. [38] found positive correlation of vitamin E and other antioxidants with TG and LDL. While our study revealed a significant negative correlation of vitamin E with TC, TG, VLDL and LDL. The above discrepancy may be because in normal population , exposure to oxidative stress results in compensatory upregulation of antioxidant system while DM as chronic condition causes depletion of the antioxidant system and also the metabolism of lipoprotein is impaired causing decrease in delivery of the fat soluble vitamin E.

Vitamin E appeared in this study to be the most important predictor of DM type II among several factors as Age, BMI, BP and different lipoproteins indicating a possibility to assess population at risk of DM type II by measuring Vitamin E. Consistently, Salonen et al. [2] in a 4-year prospective study found that men with below-median plasma vitamin E levels were associated with a 3.9-fold higher relative risk of diabetes.

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