EUROPEAN ACADEMIC RESEARCH
Vol. II, Issue 9/ December 2014

Impact Factor: 3.1 (UIF)
DRJI Value: 5.9 (B+)

ISSN 2286-4822
www.euacademic.org

Evaluation of Lipid Profile in Vitamin D Deficiency Diabetes Mellitus Type 2

MOHAMMED SIRAJELDEEN MOHAMMED
Department of Clinical Chemistry
Faculty of Medical Laboratory Science, AL-Neelain University
Sudan
AMAR MOHAMED ISMAIL[1]
Department of Biochemistry of Molecular Biology
Faculty of Science and Technology, AL-Neelain University
Sudan

Abstract:

Background: *lipid profile had major effect in many diseases worldwide especially atherosclerosis, insulin and vitamin D (vit D) play an important role on the lipids metabolism and its complications. Study aim to evaluate lipid profile level in type 2 diabetes mellitus (DM) with vit D deficiency.*

Materials and Methods: *Descriptive cross sectional study, 120 type2 DM patients aged between 25-80 years old were enrolled, then classified based on vitD results classified into two groups, (<30 ng/ml) considered as cases and (>30 ng/ml) as control groups. VitD, lipid profile (Cholesterol, triglyceride, LDL-C and HDL-C) and glucose were measured in fasting blood samples, using competitive ELISA and Mindray BS-380.*

Results *DM and vitamin D deficient are more common among females. Mean of lipid profile () showed insignificant difference when compared case with control groups, in contrast mean concentration of cholesterol significantly increased in patients who have duration >5 years, in addition no significant differences observed between others lipids and (duration of disease, BMI and gender variations).*

[1] Corresponding author: amarqqqu@yahoo.com

Conclusion *Study concludes female are more susceptible to DM and vitamin D deficient, which may lead to increase complication of vitamin D deficient among female, duration of disease effect in cholesterol levels thus patients should be monitored.*

Key words: DM diabetes mellitus, vitD vitamin D, TG triglyceride, LDL-C low density lipoprotein, HDL-C high density lipoprotein.

Introduction

Diabetes mellitus (DM) type2 is a hereditary chronic endocrine metabolic disorder, and the fifth causes of deaths worldwide [1]. This form was previously referred to as non insulin-dependent diabetes mellitus (NIDDM) or "adult-onset diabetes [2]. Type 2 DM is the most common form of diabetes, which lead to cardiovascular diseases and other complication as nephropathy and retinopathy [3,4]. Both are usually present at the time that this form of diabetes is clinically manifest, by definition the specific reasons for the development of these abnormalities are not yet known

Type 2 diabetes is associated with a cluster of interrelated plasma lipids and lipoproteins abnormalities, including reduced HDL cholesterol, a predominance of small dense LDL particles, and elevated triglycerides. These abnormalities occur in many patients despite normal LDL cholesterol levels. These changes are also a feature of the insulin resistance syndrome (also known as the metabolic syndrome), which underlies many cases of type 2 diabetes. In fact, pre-diabetic individuals often exhibit an atherogenic pattern of risk factors that includes higher levels of total cholesterol, LDL cholesterol, and triglycerides and lower levels of HDL cholesterol than individuals who do not develop diabetes. Insulin resistance has striking effects on lipoprotein size and subclass particle concentrations for VLDL, LDL, and HDL [5].

Altered vitamin D and calcium homeostasis may play a role in the development of type 2 diabetes mellitus (type 2 DM). Vitamin D and calcium insufficiency may negatively influence glycemia, whereas combined supplementation with both nutrients may be beneficial in optimizing glucose metabolism [6] , Vitamin D itself is essential for normal insulin release in response to glucose and for maintenance of glucose tolerance so that vitamin D deficiency results in decreased pancreatic insulin secretion, without altering glucagon secretion [15 ,16]. Vitamin D supplementation improves stimulated insulin secretion in response to an oral glucose load in patients with mild (normal fasting serum glucose) [17,18], type 2 diabetes mellitus, in non-diabetic healthy subjects and in subjects with vitamin D deficiency but not in patients with established type 2 diabetes mellitus [19,20]. Many researchers have confirmed that vitamin D plays an important role in endothelial function, blood pressure control, calcification of the coronary vasculature, increased vascular resistance, and prevention of CVD [21]. Vitamin D inhibits expression of uncoupling protein 2 (UCP2) in adipose tissue, differentiation of preadipocytes, synthesis and secretion of lipoprotein lipase [22].

Materials and Methods

Cross-sectional study was conducted at primary health care center (Almotakamil) at Khartoum state,120 diabetic patients type2 (aged between 25-80 years) were classified into two groups based on vitamin d level (<30ng/ml deficient, >30ng/ml control). Exclusion criteria were pregnancy, lactation, hypertension, overnight fasting, 6 ml of peripheral blood was taken. The blood samples were centrifuged at 3000 rpm for 10 min and serum stored at −20°C.utilized for different metabolic parameter, fasting blood glucose, vitamin D, lipid profile.

Ethical consideration

The study has been approved by the local ethics committee of Al-Neelain University. The study participants give their written informed consent. Sample and clinical information were used anonymously.

Measurement of BMI

Weight and height were measured and BMI was calculated by dividing weigh in (Kg) by squire of height in (m).

Estimation of vitamin D

Competitive immunoassay ELISA kit is used for quantities of vitD level, (lot E 140116AE) (EuroIMMUN AG) Germany according to the manufacture protocols. Volume of 200 ml sample diluted with biotin microplate well which coated with monoclonal anti vitD antibodies was added to wells, during incubation antigen antibodies reaction occurred, then unbounded 25-OH vitD was removal by washing process, then 100 ml of streptavidin-peroxidase was added to detect bond biotin labelled 25-OH vitamin D, furthermore 100 ml tetramethylbenzidine which promotes colour reaction, the colour intensity developed is inversely proportional vitD concentration in the sample, which calculated by blotting standards curve by using (Sunrise-TECAN) ELISA reader [7,8].

Estimation of Glucose

Glucose present in the plasma oxidized by glucose oxidase to hydrogen peroxide, which react with 4-aminoantipyrine and phenol to form quinoneimine (pink color), which absorbed at 520±20 nm using spectrophotometer BTS-310 Biosystem, the concentration obtained by calculation of optical density of test against optical density of standard, 10 µl of serum added to 1ml of reagent incubated for 10 min [9].

Estimation of lipid profile

TG: glycerokinase peroxidise peroxidase method, through sequences of enzymatic catalysis steps by lipase G.K and G.P.P. triglyceride is catalysed yield H2O2 which oxidised for amino anti pyrinel to yield colour dye of quinonimine, the absorbance increase directly proportional to the concentration of T.G.

Procedure: By the Mindray BS380 to 1ml of reagent add 10microlier of sample; mix well and read the absorbance after incubation at 37 0C [10].

T.C: Cholesterol oxidase peroxidase method. By the catalysis of CHE and CHO cholesterol ester is catalysed to yield H2O2 which oxidised for amino anti-pyrine with phenol to form colour dye of quinonimine; the absorbance increase directly proportional to concentration of cholesterol.

Procedure: By the mindray BS380 to 1ml of reagent add 10 µl of sample; mix well and read the absorbance after incubation at 37 0C [11].

LDL, HDL: Direct method: The system monitors the change in absorbance at 600 nm; this change directly proportional to the concentration of cholesterol in the sample and used by the system to calculate and expressed the LDL and HDL cholesterol [12,13].

Procedure: By the Mindray BS380 to 900 µl of reagent 1 add 12microlier of sample; mix well and incubate at 37 0C for 5 min; then add 300 µl of R2, then mixed well and incubate at 37 ^{o}C for 5 min then read the absorbance.

Statistical analysis

Data from all patients were presented as (Mean±SD), differences between means of patients and control groups were considered statistically significant with p-value threshold <0.05 using independent T-test.

Results

Table.1. Showed lipid profile among case (vitamin D3 deficient patients) and control (normal vitamin D3) (n=120)

Variables	Case (Mean±SD)	Control (Mean±SD)	*P*-value
Cholesterol	(191±38)	(189±44)	0.849
Triglyceride	(136±73)	(157±90)	0.161
LDL-C	(118±30)	(114±35)	0.473
HDL-C	(41±7.6)	(34±8.2)	0.371

Table.2. Revealed vitD and lipid profile among type2 DM males and females (n=120)

Variables	Male (Mean±SD)	Female (Mean±SD)	*P*-value
Vitamin D3	(30±15.7)	(24±12.6)	0.032
Cholesterol	(195±43.9)	(187±38.6)	0.311
Triglyceride	(153±92)	(137±72)	0.281
LDL-C	(122±35.1)	(114±29.7)	0.148
HDL-C	(41.9±8.2)	(41.8±7.4)	0.965

Table.3. Revealed vitD and lipid profile among type2 DM according to BMI (n=120)

Variables	>26.5 (mean±SD)	<26.5 (mean±SD)	P. value
Vitamin D3	(25.8±12.8)	(28.7±16.9)	0.390
Cholesterol	(189±35.5)	(194±52.2)	0.550
Triglyceride	(141±77.4)	(148±87.9)	0.665
LDL-C	(115±29.5)	(121±37.6)	0.361
HDL-C	(41.7±7.7)	(42.3±7.8)	0.786

Table 4. Revealed vit D and lipid profile among type2 DM according to duration of disease (n=120)

Variables	>5 years (mean±SD)	<5 years (mean±SD)	P. value
Vitamin D3	(27.3±15.3)	(25.8±12.4)	0.565
Cholesterol	(197±42.6)	(181±36.5)	0.035
Triglyceride	(153±88.7)	(131±66.9)	0.120
LDL-C	(120±35.1)	(113±27.5)	0.261
HDL-C	(42.6±7.6)	(41.1±7.8)	0.405

Discussions

DM is a chronic disease, worldwide results from a lack of insulin or inadequate insulin secretion following increases in insulin resistance. Abnormal lipid profile and lipoproteins oxidation especially LDL-C is more common in diabetic patients and are aggravated with a poor glycaemic control [14]. The study aims to evaluate lipid profile level among type2 DM patients with vitamin D deficiency.

Twenty hundred DM subjects were enrolled in this study. DM is higher among females 76 (63.0%) than males 44 (37.0%), in addition vitD deficiency is more common among female than male with (*p*-value 0.032), our findings agreed with previous report that stated the prevalence of type 2 DM is higher among females than males, also found that vitamin D is decreased in females, this observation indicate that female more susceptible to DM and vitD deficiency than male which may due to the nature of males work more exposure to the sun light than females, factor for vitamin D synthesis and thus prevent beta-cells damage consequently reduce DM among male [23,24 , 26].

The results of lipids showed that, there were insignificant differences in mean (cholesterol ,TG , LDL-C and HDL-C) levels of patients in comparison with control group with (*P*-value 0.849 ,0.161 ,0.473,0.371) respectively, in fact that in type 2 DM patients with vitamin D deficiency serum level of cholesterol, triglycerides, LDL-C were higher and HDL-C was lower compared to normal vitamin D patients, with only statistically significance in triglycerides, our finding revealed the same results with previous study, which may attributed to effect of DM on lipids metabolism as in control group, in spite of the control has normal vitamin D but also has uncontrolled DM [25].

The results showed insignificant difference in mean vitD levels of overweight patients compared with normal weight DM

patients with (*p*-value 0.390) which agreed with previous study that found there was insignificant difference in vitamin D level of overweight DM patient in comparison with normal weight [22]. In addition the results of lipids revealed that, insignificant differences were observed in mean concentration of (cholesterol, TG, LDL-C and HDL-C) of overweight DM patients when compared with normal weight with *P*-values (0.565, 0.390 ,0.550 ,0.665 0.361) respectively. Which Agreed with previous study in cholesterol finding and in conflict in other lipids parameters (TG, LDL-C and HDL-C) [28].

The results of present study provide experimental evidence that, there were insignificant differences between mean of cholesterol, TG, LDL-C and HDL-C levels in males when compared with females DM patients with *P*-value (0.311, 0.281, 0.148, 0.965) respectively, agreed with previous study that found insignificant differences in lipid profile among gender variation [29].

Significant increase in mean cholesterol level was observed in patients who have >5 years type 2 DM in comparison with those have <5 years duration of disease with (*P*-value 0.035), in contrast the results of others lipids showed insignificant differences among groups classified based on duration of disease, which calcified by previous study, that presence of high prevalence of dyslipidemia (hypercholesterolemia) in type 2 diabetic patients [27].

Conclusion:

Study concludes female are more susceptible to DM and vitamin D deficient than male, which may increase the risk of complication DM among females, duration of DM >5 years significantly increased cholesterol level thus may lead to hypercholesterolemia and its complication.

REFERENCES:

[1]Faghilimnai S, Hashemipour M, Kelishadi B. The lipid profile of children with type 1 diabetes as compared to the controls. *ARYA. J* 2006; 2(1):36-38.

[2] Melissa Conrad Stoppler., diabetes mellitus. *Medicine Net* (2014), *14*; 2.

[3] Miller M. The epidemiology of triglycerides as a coronary artery disease risk factor *Clin. Cardiol* 1999; 22 (II):111-113.

[4] Frohlich J, Dobiasova M, Lear S, Lee KWJ:The role of risk factors in the development of atherosclerosis. *Crit Rev Cl Lab Sci* 2001,38(5):401–440.

[5]Rosenson, R.S.: HDL-C and the diabetic patient: target for therapeutic intervention. Diabetes Res. *Clin. Diabetes Res. Clin. Pract*. ,(2005) : 68(2) : 36-42 .

[6] Pitter M H, C S Stevenson, E.Ernst., Chromium piclonate for reducing body weight:Metea analysis of randomized trials. *International Journal of Obesity*.(2003) 27; 522-529.

[7] Holick M F, Chen T C.Vitamin D deficiency: worldwide problem with health consequence.*American Clinical Nutrition*. (2008), 87; 1080-1086.

[8] Hollis Editorial., The Determination of circulating 25-Hydroxyvitamin D: No Easy Task BW *J ClinEndocrinolMetab*. (2004), 89(7); 3149-3151.

[9] Trinder P., Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Ann Clinical Biochemistry* (1969), 6; 24-27.

[10] Thomas l.clinical laboratory Diagnostics 1st ed.frankfurt: TH books veriagsgesellschaft; 1998.

[11] Recommendation of the second joint task force of European and other societies on coronary Prevention of coronary heart disease in clincial EUr heart j 1998 19: 1434-503.

[12] Tietz textbook of clinical chemistry 3rd edition .Burtis CA, AshwoodER, WB Saunders CO, 1999.

[13] Tietz textbook of clinical chemistry 3rd edition .Burtis CA, AshwoodER, WB Saunders CO, 1999.

[14]Monouk Bos, Charlis Agyemang, Prevalence and complication of DM in northern Africa. *BMC public health 2013*.13; 1-7.

[15] Ismail A, Namala R. Impaired glucose tolerance in vitamin D deficiency can be corrected by calcium. *J Nutr Biochem 2000*; 11: 170–175.

[16]Kadowaki S, Norman AW. Dietary vitamin D is essential for normal insulin secretion from the perfused rat pancreas. *J Clin Invest* 1984; 73: 759–766.

[17]Chiu KC, Chu A, Go VLW et al. Hypovitaminosis D is associated with insulin resistance and ß cell dysfunction. *Am J Clin Nutr 2004*; 79: 820–825.

[18]Inomata S, Kadowaki S, Yamatani T et al. Effect of 1α(OH)-vitamin D_3 on insulin secretion in diabetes mellitus. *Bone Miner 1986*; 1: 187–192.

[19]Orwoll E, Riddle M, Prince M. Effects of vitamin D on insulin and glucagon secretion in non-insulin-dependent diabetes mellitus. *Am J Clin Nutr 1994*; 59: 1083–1087.

[20]Gedik A, Akalin S. Effects of vitamin D deficiency and repletion on insulin and glucagon secretion in man. *Diabetologia 1986*; 29: 142–145.

[21] Nemerovski CW, Dorsch MP, Simpson RU, Bone HG, Aaronson KD, Bleske BE: Vitamin D and cardiovascular disease. *J Hum Pharmacol Drug Ther* 2009, **29**(6)**:**691-708

[22] Ehsaneh Taheri, Ahmad Saedisomeolia, Mahmoud Djalali, Mostafa Qorbani, and Manouchehr Madani Civi . The relationship between serum 25-hydroxy vitamin D concentration and obesity in type 2 diabetic patients and healthy subjects. *J Diabetes Metab Disord*. 2012; 11: 16

[23] P. Lips, D. Hosking, K. Lippuner, J. M. Norquist, L. Wehren, G. Maalouf, S. Ragi-Eis and J. Chandler. The prevalence of vitamin D inadequacy amongst women with

osteoporosis: an international epidemiological investigation, *J Internal Medicine2006*, 260, 3 :245–254 .

[24]Mansour Alzahrani, Prevalence of vitamin d deficiency in type 2 DM, *J. HEALTH SCIENCES*, 2013 –v1 e 324

[25] Ahmad Saedisomeolia, Ehsaneh Taheri, Mahmoud Djalali, Ali Malekshahi Moghadam and Mostafa Qorbani. Association between serum level of vitamin D and lipid profiles in type 2 diabetic patients in Iran, *Journal of Diabetes & Metabolic Disorders* 2014, 13:7

[26] Marina Scavini, Christine A, Vallabho, Andrew S, Francesca T, David S, Arlene B, Carleton P, Jeanette B, Eunice J, Kathy N, Donica N, Mildred W, Thomas K, Jean W and Philip G, Prevalence of diabetes is higher among female than male Zuni Indian. *American diabetes association Diabetes care*. (2014), 26; 55-60.

[27] M. Nakhjavani A. R. Esteghamati , F. Esfahanian and A. R. Heshmat, Dyslipidemia in Type 2 Diabetes Mellitus: More Atherogenic Lipid Profile in Women. *Acta Medica Iranica*, 2006, 44(2): 111-118.

[28] Ratna Manjula Songa, Siddhartha, K., Sudhakar, K. Lipid profile in type 2 DM with obesity, *Bulletin of pharmaceutical and medical sciences (BOPAMS*), 2013, v1, e 2 .

[29]Sayed shahid, Muhammad Aslam, Wagas Hameed, gender differences in lipid and lipoproteins (a) profiles in healthy individuals and patients with type 2 diabetis mellitus ,*Pak J physiol* 2005, 1,(1-2).