

STUDY OF HbA1c, LIPID PROFILE AND CYCLOPHILIN-A IN DIABETES MELLITUS

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Abstract

Introduction: Diabetes is a chronic disease that occurs when not enough insulin is produced by the pancreas or the body does not use the insulin produced. Because of increased blood glucose levels in the body, serious heart, kidneys, blood vessels, nerves and eyes damage are caused. Report says about 400 million people suffer from diabetes. Therefore present study is aimed to assess levels of HbA1c, Lipid profile and Cyclophilin A in diabetic patient.

Material and Methods: The present study includes total 126 subjects comprising of 66 type 2 Diabetes Mellitus patients and 60 healthy individual. Blood samples are collected from the all subjects were processed for HbA1c, Lipid Profile and Cyclophilin A estimation, from OPD and General Medicine Wards. HbA1c is estimated by HPLC, lipid Profile by AU480 and the Cyclophilin A by ELISA method using commercially available Qayee-bio ELISA kit.

Conclusion: Present study showed significantly increased levels of HbA1c, Lipid Profile and Cyclophilin A in T2DM patients. The elevated lipid profile may be due to the complication of Diabetic mellitus. CyA is increased as an inflammation marker.

Keywords: T2DM: Type 2 diabetes mellitus, HbA1c: Glycosylated Hemoglobin, CyA: Cyclophilin-A

Introduction

Diabetes is a group of metabolic diseases that are characterized by hyperglycaemia caused by insulin secretion defects, insulin action, or both. Chronic diabetes hyperglycaemia is associated with long-term damage, dysfunction, and failure of various organs, particularly the eyes, kidneys, nerves, heart, and blood vessels. ⁽¹⁾

Type-1 DM results from the failure of the body to produce insulin, and the person is required to be on insulin treatment. This usually happens in children or young adults, but can happen at any age (Joshin Diabetes Center, 2007) ⁽²⁾. Type-2 DM (formerly known as adult onset) results from insulin resistance, a condition in which cells fail to properly use insulin, sometimes combined with an absolute deficiency in insulin. Formerly Referred to As Non-Insulin-Dependent Diabetes Mellitus (NIDDM) Or Adult-Onset Diabetes ⁽³⁾.

The clinical usefulness of HbA1c as a tool for assessing the risk of diabetes complications was suggested when publishing the results of DCCT46 and UKPDS47 these studies established the effect of intensive (as compared to conventional) glycemic

control on the development of micro vascular complications in patients with type 1 and type 2 DM respectively. ⁽⁴⁾

Lipid abnormalities associated with diabetes are termed as dyslipidemia rather than hyperlipidemia. Diabetes mellitus (DM) is a common secondary cause of hyperlipidemia, particularly, if glycemic control is poor, which in-turn is an important risk factor for atherosclerosis and coronary heart disease. ⁽⁵⁾

Cyc-A is a highly abundant and cytosolic protein. The protein belongs to a family of isozymes, including cyclophilins - B and C, and the protein associated with the natural killer cell cyclophilin. Major Isoforms, including the ER, were found throughout the cell, and some are even secreted. Cyc-A is also known as peptidylprolyl isomerase A, found in the cytosol, contains a beta barrel structure with two alpha helices and a beta sheet. Other cyclophilins have structures like cyc-A. Cyclosporine cyclophilin - A complex inhibits calcium/calmodulin - dependent phosphatase, calcineurin, inhibiting organ rejection by stopping the production of TNF - alpha and interleukin-2 pro - inflammatory molecules. ^(6,7)

The secretory nature of this protein and its presence in the plasma of diabetes mellitus and CAD patients

underscores its potential as a disease marker. Secretion of cyc-A in hyperglycemic conditions from monocytes. Preliminary studies of a small number of patients with type-2 diabetes mellitus and coronary artery disease showed an increase in cyc-A Plasma levels compared to normal healthy individuals. ^(8,9)

MATERIAL AND METHODS:

The study is conducted in the Department of Biochemistry in collaboration with Department of General Medicine at MGM Medical College Mumbai. T-2 DM Patients in Group-1 are enrolled from Department of General Medicine ward, Diabetic OPD and for healthy individuals Group-2 from general population as well as relatives of patients in Medicine OPD. Total 126 subjects were enrolled and grouped as mentioned.

After 12 hours overnight fasting, 5 ml of blood is collected from each subject by venepuncture with standard blood collection technique in a plain vial for serum separation, sodium fluoride vial for plasma and EDTA vial for HbA1c estimation. HbA1c estimation was done by using HPLC method ⁽¹⁰⁾. Plasma was collected after two hours of post meal for the postprandial glucose estimation. Estimation of Plasma glucose done by using GOD-POD method ⁽¹¹⁾. Serum is used for lipid profile (TC, TG and HDL) ⁽¹²⁻¹⁴⁾. Samples were stored at -70°C for estimation of CypA. Serum Cyclophilin A is estimated by ELISA method using commercially available Qayee-bio ELISA kit ⁽¹⁵⁾.

RESULT:

Graph 1: Showing Age group wise distribution of T2DM and Healthy Control

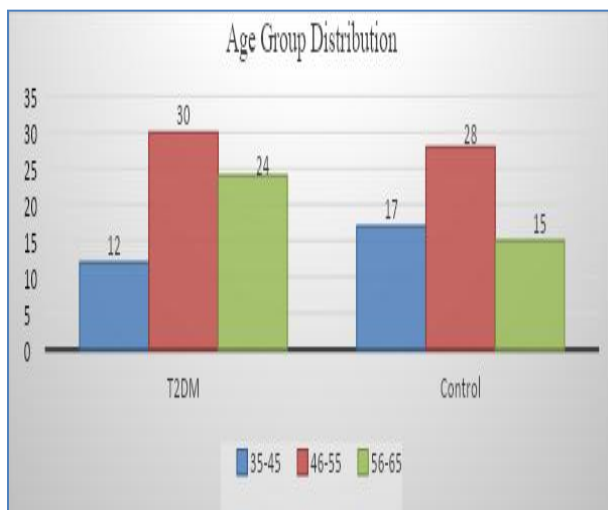


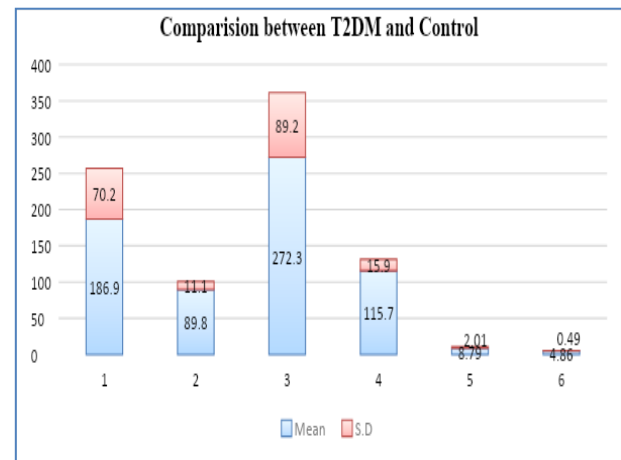
Table 1: Showing age wise distribution of T-2DM and Healthy Control subjects.

Age (Years)	T2DM	Healthy Control	Total Subject
35-45	12 (10%)	17 (13%)	29 (23%)
46-55	30 (24%)	28 (22%)	58 (46%)
56-65	24 (18%)	15 (13%)	39 (31%)
Total	66 (52%)	60 (48%)	126 (100%)

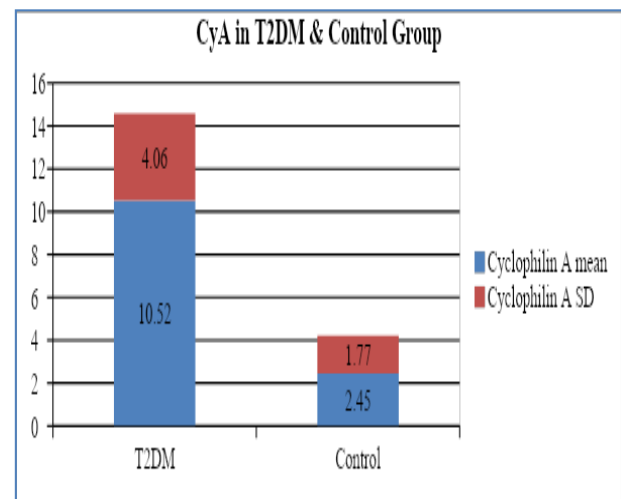
Table 2: Mean FBS, PPBS, HbA1c and Cyclophilin A in T2DM and Control Group

Biochemical Parameters	Diabetic group	Healthy control	P value
FBS (mg/dl)	186.9±70.2	89.8±11.1	<0.0001
PPBS (mg/dl)	272.3±89.2	115.7±15.9	<0.0001
HbA1c (%)	8.79±2.01	4.86±0.49	<0.0001
Cyclophilin-A (ng/ml)	10.52±4.06	2.45±1.77	<0.0001

P<0.0001, statistically significant



Graph 2: Mean FBS, PPBS and HbA1c amongst T2DM and Control Group

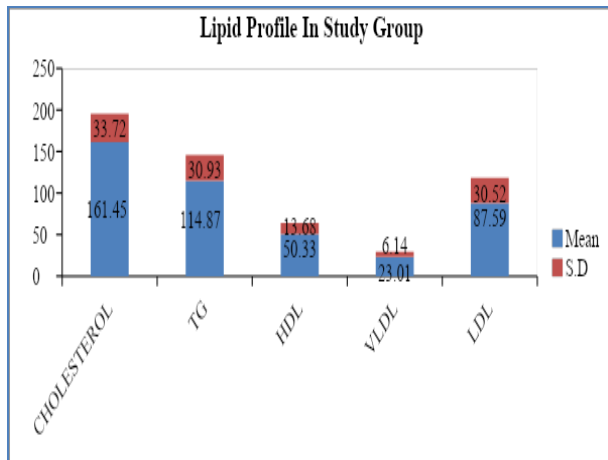
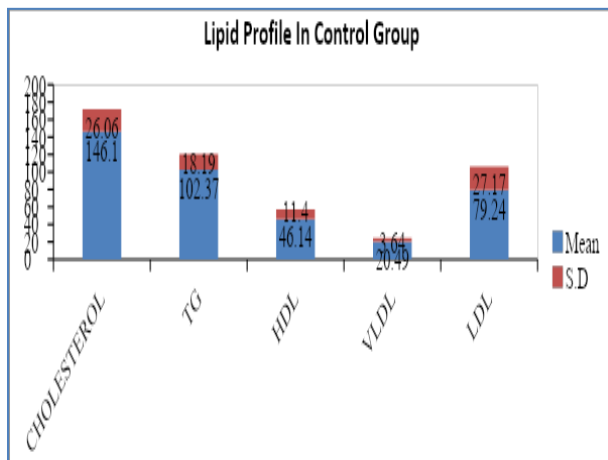


Graph 3: Mean Cyclophilin A amongst T2DM and Control Group

Table 3: Mean lipid profile amongst T2DM and Control Group

	Diabetic Group	Healthy Control	P value
Cholesterol (mg/dl)	161.45±33.72	146.1±26.06	0.0053**
Triglyceride (mg/dl)	114.87±30.93	102.37±18.19	0.0073**
HDL (mg/dl)	50.33±13.68	46.14±11.40	0.0656 [@]
VLDL (mg/dl)	23.01±6.14	20.49±3.64	0.0065**
LDL (mg/dl)	87.59±30.52	79.24±27.17	0.1087 [@]

** Statistically significant, @ not statistically significant

**Graph 4:** Mean lipid profile amongst T2DM Group**Graph 5:** Mean lipid profile amongst Control Group

DISCUSSION

With more than 62 million diabetics currently diagnosed with disease, diabetes is rapidly gaining the status of a potential epidemic in India. India's diabetes etiology is multifactorial, involving genetic factors coupled with environmental influences such as rising living standards, steady urban migration, and lifestyle changes ⁽¹⁶⁾.

Present study is conducted in MGM Medical College; patients are selected from Diabetic OPD. The subjects included in present study are informed about study and written consent is obtained before sample collection from the study group & healthy control. The study includes total 126 subjects, including Group I (66 T-2DM subjects) and Group II (60 Healthy individuals).

As mentioned in Table I and Graph I, maximum number of subjects are from 46-55 years (46%) followed by 56-65 years (31%) and 35-45 years (23%).

Table 2 and graph 2 shows, mean FBS is significantly higher in T-2DM patient (186.2±70.2) mg/dl as compared to healthy individual (89.8±11.1) mg/dl, mean PPBS is significantly higher in Type-2 Diabetes Patient (272.3±89.2) mg/dl as compare to control (115.7±15.9) mg/dl and mean HbA1c is significantly higher in T-2DM patients (8.79±2.01) % as compared to control group (4.86±0.49) %. (p<0.0001)

Further table 2 and graph 3 shows, mean Cyclophilin A is significantly higher in T-2DM patient (10.52±4.06) ng/ml as compared to healthy individual (2.45±1.77) ng/ml. Serum Cyclophilin A is significantly higher in T-2DM patient as compared to healthy individual. (P<0.0001). Our Results are concurrent with Ramchandran et al 2014, he reported that in patients with diabetes and coronary artery disease plasma cyclophilin levels were increased suggesting a role for this protein in accelerating vascular disease in type 2 diabetes. Considering the evidence that Cyclophilin A in thermogenesis is an inflammatory mediator, the mechanical role of Cyclophilin A in diabetic vascular disease. ⁽¹⁷⁾

Table 4 shows the mean level of lipid profile in Group 1 and Group 2 i.e. Study Group and Control group respectively. Total cholesterol levels are very significantly increased in Group I as compared to Group II (P<0.0053). Triglyceride levels are highly increased in study group (i.e. group I) as compared to group II (P<0.0073). Level of VLDL is very significantly increased in study group than in control group (P<0.0065).

Table 3 and graph 4 and 5 shows, mean Cholesterol is significantly higher in T-2DM patient (161.45±33.72 mg/dl) as compared to healthy individual (146.1±26.06 mg/dl) (p=0.0053), the mean Triglyceride is significantly higher in Type-2 Diabetes Patient (114.87±30.93 mg/dl) as compare to control

(102.37±18.19mg/dl) (p=0.0073) and the mean HDL is non-significantly higher in T-2DM patients (50.33±13.68 mg/dl) as compared to control group (46.14±11.40 mg/dl) (P<0.0656). The mean VLDL is significantly higher in Type-2 Diabetes Patient (23.01±6.14 mg/dl) as compare to control (20.49±3.64) mg/dl (p=0.0065) and the mean LDL is non-significantly higher in T2DM patients (87.59±30.52 mg/dl) as compared to control group (79.24±27.17 mg/dl) (P=0.1087).

Diabetes is associated with a higher risk of cardiovascular disease (CVD) mortality, known as dyslipidemia, characterized by elevated triglycerides, low-density lipoprotein and high-density low-density lipoprotein particles. It may be present in the diagnosis of type-2 diabetes mellitus and is a metabolic syndrome component. Abnormal serum lipids in diabetic patients are likely to contribute to the risk of coronary artery disease. Anomalies in lipids are common in diabetics and often seen in diabetics of type 2. Dyslipidemia makes diabetics prone to heart disease (CHD and other atherosclerosis complications). Our result concurrent with Sultania S et al⁽¹⁸⁾, Bhambhani GD et al⁽¹⁹⁾ & Biradar SB et al⁽²⁰⁾.

Sultania S et al. 2017⁽¹⁸⁾ demonstrated the typical low HDL, high triglyceride diabetic dyslipidemia. This pattern of dyslipidemia has also been demonstrated by various national and international epidemiological studies on lipid profile. In this study, there was no significant difference in total cholesterol and absolute LDL levels in cases and checks. Even if the absolute LDL cholesterol concentration (LDL-C) has not increased significantly.

In their study, Bhambhani GD et al. 2015⁽¹⁹⁾ showed that the majority of patients with type 2 DM (72%) had high serum cholesterol levels, whereas only 12% of patients with type 1 DM had high serum cholesterol levels. Serum TG has been raised in 48% of patients with type 2 DM. In our study, the values of serum TG are consistent with the above study. The reason why the serum cholesterol values differ may be due to differences in people's dietary habits.

Biradar SB et al. 2018⁽²⁰⁾ the study T2DM patients showed significant increases in TGL and VLDL serum, further the study showed increases in TC and LDL levels that were not statistically significant compared to normal healthy control group. T2DM patients experienced a significant decrease in HDL compared to the normal control group.

In the current study we found significant increase in Cholesterol, triglyceride and VLDL similar as per previous studies. In present study we found Non-significant increase in HDL and LDL level.

CONCLUSION

The objective of this is to evaluate Cyclophilin A, HbA1c and Lipid profile in diagnosed cases of type-2 diabetes patients and compare with healthy control. Cyclophilin A are significantly elevated in type-2 diabetes patients. These biomarkers have a critical role in diagnostic, therapeutic and prognostic decision-making, particularly in the context of inadequate quantitative risk assessment available for clinicians. These parameters will help to diagnose Type 2 Diabetes, its severity, complications and metabolism in detail. This could significantly decrease the burden of complications imposed on patients and the healthcare system via planning to reduce modifiable risk factors.

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