Original article:

Lipoprotein (a), C-Reactive protein and serum uric acid as cardiovascular risk factors in type 2 diabetes mellitus

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Abstract

Diabetes mellitus is a disorder of carbohydrate metabolism leading to hyperglycemia and are at increased risk of its complications. Higher morbidity and mortality is due to its cardiovascular complications. This study is to assess the role of non- traditional factors like Lipoprotein(a), Highly sensitive C Reactive Protein(Hs-CRP), Uric acid for the cardiovascular risk in type 2 Diabetic patients. Methods: Fasting Blood Sugar(FBS), Lipid profile, Lipoprotein (a), Hs-CRP and Uric acid were estimated in age and gender matched 60 type 2 diabetic individuals as cases and 60 healthy individuals as controls. Results: Levels of Lipoprotein (a), Hs-CRP, Uric acid, Total Cholesterol, Triacylglycerol and LDL-Cholesterol were significantly elevated in diabetic individuals. Conclusion: The parameters under study have been significant in assessing the cardiovascular risk in diabetic individuals.

Key words: Lipoprotein(a), Hs-CRP, Uric acid, Type 2 Diabetes mellitus

Introduction

The prevalence of diabetes worldwide was estimated to be 4% in 1995 and anticipated to rise to 5.4% by 2025 [1]. Currently India has 40.9 million people with diabetes and the projected estimate for the year 2025 is 69.9 million[2]. The morbidity and mortality is higher in patients with type 2 diabetes due to cardiovascular complications and the risk of atherosclerotic coronary artery disease is increased by 2-4 fold in type 2 diabetics[3]. The traditional risk factors do not fully explain this scenario. In this regard it would be of great interest to assess the role of nontraditional factors such as Lipoprotein (a), C-reactive protein and Uric acid for cardiovascular risk in diabetic patients[4].

Material and methods

A Case control study was conducted during Aug2011-Sep 2012, in the Department of Clinical Biochemistry, Osmania General Hospital, Hyderabad; Andhra Pradesh. The subjects were divided into 60 cases and 60 controls. Cases included Type II Diabetic patients in the age group of 40-60 yrs of both genders and Controls comprised of Non diabetic healthy individuals not suffering from any ailments. Individuals with Fasting Blood Sugar above 110 mg/dl were considered to be diabetic and those with FBS between 70-110 mg/dl were considered to be non diabetic.

Exclusion Criteria:

1. Type I diabetic individuals
2. Individuals with history of Tuberculosis, hepatic diseases, Typhoid, Epilepsy, Stroke, Asthma, Allergies, Anaemias, Malignancies
3. Individuals with history of alcoholism and smoking
4. Individuals with Severe Combined Immunodeficiencies

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5. Individuals on treatment with statins, insulin, thiazide diuretics, hyperuricemic drugs and multivitamins
6. Pregnant women, Gestational diabetics, Women on Hormone Replacement Therapy.

Participants were in overnight fasting status. 5ml of venous blood drawn from all the cases and controls with their consent. Plasma and serum separated by centrifuge within one hour of sample collection. Plasma is used to analyze Fasting Blood Sugar(FBS) by Glucose Oxidase Peroxidase method. Serum is used to analyze Lp(a) by Latex Turbidimetry, Hs-CRP by Immunoturbidimetric method, Uric acid by Uricase method. In Lipid profile, Total Cholesterol by Cholesterol oxidase peroxidase method, HDL-C by Precipitation End Point method, TAG by Glycerol Phosphate Oxidase Peroxidase method and LDL-C, VLDL-C values calculated.

The collected data was analyzed using SPSS (statistical package for social science version 17.0) statistical package. Descriptive Statistics like Range, Mean and Standard Deviation (SD) were computed for each biochemical parameter for case as well as control group. Student's t-test was used for testing the significance difference in mean scores of various bio-chemical parameters between case and control groups

Results :

MEAN±SD, t-VALUES OF PARAMETERS IN CASES AND CONTROLS

Table: 1

Parameter		N	Mean	SD	t-value
FBS	Cases	60	167.63	31.513	18.261**
	Controls	60	89.48	10.290	
LP(a)	Cases	60	56.327	15.820	19.012**
	Controls	60	14.617	6.1454	
Hs-CRP	Cases	60	6.38	1.774	18.267**
	Controls	60	1.75	.841	
URIC ACID	Cases	60	7.422	.7223	26.68**
	Controls	60	3.218	.3270	
T.CHOL.	Cases	60	214.98	33.935	10.231**
	Controls	60	161.88	21.553	
HDL-C	Cases	60	35.50	5.104	7.465**
	Controls	60	81.90	47.873	
TAG	Cases	60	223.08	48.449	18.964**
	Controls	60	91.46	23.301	
LDL-C	Cases	60	134.213	28.3294	10.731**
	Controls	60	84.117	22.4765	
VLDL-C	Cases	60	44.923	10.0222	18.037**
	Controls	60	17.770	4.8826	

***- p<0.01, Statistically Highly Significant; NS- p>0.05, not significant*

The Mean (±SD) of FBS in Cases and Controls is 167.63(31.513) and 89.48(10.290) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

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The Mean (±SD) of Lp(a) in Cases and Controls is 56.327(15.820) and 14.617(6.145) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

The Mean (±SD) of Hs-CRP in Cases and Controls is 6.38(1.774) and 1.75(.841) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

The Mean (±SD) of Uric acid in Cases and Controls is7.422(.7223) and 3.218(.3270) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

The Mean (±SD) of Total Cholesterol in Cases and Controls is 214.98(33.935) and 161.88(21.553) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

The Mean (±SD) of HDL-C in Cases and Controls is 35.50(5.104) and 81.90(47.873) respectively and the difference in the mean was found to be significantly lower in diabetics than controls ($p<0.01$).

The Mean (±SD) of TAG in Cases and Controls is 223.08(48.449) and91.46(23.301) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

The Mean (±SD) of LDL-C in Cases and Controls is 134.213(28.3294) and 84.117(22.4765) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

The Mean (±SD) of VLDL-C in Cases and Controls is 44.923(10.0222) and17.770(4.8826) respectively and the difference in the mean was found to be significantly higher in diabetics than controls ($p<0.01$).

Discussion

Diabetes mellitus is the most common endocrinological disorder which is characterized by metabolic abnormalities and long term complications[5]. Long term cardiovascular complications represent the main cause of morbidity and mortality in diabetic patients[7]. Population based studies of cardiovascular disease risk among type2 diabetic and non diabetic subjects were conducted, showing increased risk in diabetic subjects[8]. Traditional risk factor like dyslipidemia was studied extensively among diabetic subjects in the previous studies. Studies on the risk factors which increase the prognostic efficiency for cardiovascular disease risk in diabetics subjects are very few. This study is conducted to find the association of novel risk factors like Lipoprotein(a), Highly sensitive C reactive protein, Uric acid with cardiovascular disease in type2 diabetic subjects.

Dyslipidemia management is especially important for those with type 2 diabetes, because type 2 is associated with a two to fourfold excess risk of coronary heart disease (CHD)[6]. Dyslipidemia management is very important and optimal levels of various lipid should be,

LDL cholesterol levels: <100 mg/dl

HDL cholesterol levels: >45 mg/dl

Triacylglycerol levels**:** <200 mg/dl

In the present study, the diabetic individuals had increased levels of Total Cholesterol(214.9±33.9),LDLCholesterol(134.2±28.3),VLDLCholesterol(44.9±10.0),Triacylglycerols (223±48.4) anddecrease in HDL-Cholesterol (35.5±5.1), compared to healthy nondiabetic individuals with TotalCholesterol(161.8±21.5),LDLCholesterol(84.1±22.4),VLDLCholesterol(17.7±4.8),Triacylglycerol(91.4±23.3) and increase in HDL-Cholesterol(81.9±17), showing the correlation of dyslipidemia with cardiovascular disease risk in diabetics. Ramirez and Co-workers[9] reported that poorly controlled diabetes mellitus is associated with a high Lp(a) level and also suggested that this metabolic abnormality contributes to the elevated coronary risk in diabetic persons. However, the mechanism of increased Lp(a) levels in poorly controlled diabetics is not clear. According to a hypothesis there exists a defect in the clearance of the apoprotein B-l00 in diabetic persons. On the

other hand, a decrease in LDL in cellular metabolism in diabetes mellitus is proposed to be due to glycation of the LDL particle and the LDL receptor. The studies by Morishita et al[10] showed significantly elevated levels of Lp(a) in patients with NIDDM. The present study also showed increased levels of lipoprotein(a) in type2 diabetics with higher fasting plasma glucose levels and that these people are at increased risk of cardiovascular disease in the future. . Heller et al (1993)[11] suggested that hyperinsulinaemia could be a causal factor for the increase in the Lp(a) levels in T2DM. Wolffenbuttel et al (1993)[12] also reported that the Lp(a) levels were elevated in diabetics as compared to the non-diabetic subjects of similar age. Coppola et al. have shown increase of circulating acute phase proteins in type 2 diabetes[13]. There are several possible mechanisms by which diabetes might induce inflammation state. In hyperglycaemic condition the concentration of advanced glycation end products is elevated that have been shown to activate macrophages, increase oxidative stress and upregulate the synthesis of IL-1, IL-6 and TNF, resulting in the production of CRP. Another possibility is that increases in CRP concentrations are related to adipose tissue derived cytokines[13]. In another study Soinio et al. have reported that in subjects with type 2 diabetes CRP is a predictor for coronary heart disease deaths[14].

In the present study, Hs-CRP levels were found to be raised in type2 diabetic cases(6.3±1.7) compared to non-diabetic controls(1.7±0.8).

Serum uric acid reflex pre-diabetic status at renal level. Hyperinsulinemia due to insulin resistance modify handling of uric acid by kidney. Increased activity of HMP shunt linked to insulin resistance cause increase in uric acid level**.** For 1mg/dl increase the CVD risk in type2 diabetics increases by 15-20%**.** It Inhibits NO production by inducing endothelial dysfunction suggesting increased levels of uric acid are not only due to insulin resistance but it promotes insulin resistance. Hyperuricaemia has been presumed to be a consequence of insulin resistance rather than its precursor and has been presumed to be associated with oxidative stress to be related to the development of the complications in diabetes[15]. In the present study uric acid levels were found to be raised in type2 diabetic subjects (7.4±0.7) compared to controls (3.2±.03). The novel risk factors like Lipoprotein (a), Hs-CRP, Uric acid were studied in the present study and found to have significant positive association in type2 diabetics at risk of cardiovascular disease.

Summary and conclusion

The morbidity and mortality due to cardiovascular complications is higher in type 2 diabetes. Studies on the risk factors which increase the prognostic efficiency for cardiovascular risk in diabetic subjects are very few. This study was conducted to find the association of novel risk factors like Lipoprotein(a), Highly sensitive C reactive protein, Uric acid with cardiovascular disease in type2 diabetic subjects. In this study, there is increase in Total Cholesterol, LDL-Cholesterol, VLDL-Cholesterol, Triacylglycerol and decrease in HDL-Cholesterol in diabetic cases compared to non diabetics indicating atherosclerotic changes in diabetics.

In diabetic patients with advanced CVD, Lp(a) indicates a coagulant risk of plaque thrombosis. Apo (a) contains domains that are similar to plasminogen. Lp(a) accumulates in the vessel wall and inhibits the binding of plasminogen to the cell surface reducing plasmin generation which increases clotting. This inhibition of plasminogen by Lp(a) also promotes proliferation of smooth muscle cells. These unique features of Lp(a) suggests that Lp(a) causes generation of clots and atherosclerosis[16].

Lipoprotein(a) concentration were found to be raised in the type2 diabetic subjects compared to non diabetics. Findings of the present study suggest that the assessment of lipoprotein(a) concentration could contribute to the identification of diabetic patients with high risk of death due to CVD. Systemic inflammatory activity has turned

out to play a key pathogenic role in vascular atherosclerosis, insulin resistance, and type2 diabetes mellitus. Inflammatory biomarkers may therefore be a valuable tool for risk evaluation. Among them, the best evidence to date supports the use of highly-sensitive C-reactive protein (Hs-CRP) to monitor insulin resistance and cardiovascular risk in diabetic and non diabetic individuals. The levels of highly sensitive C-reactive protein were found to be raised in the diabetic subjects suggesting its role as a cardiovascular risk factor in these subjects. Studies were demonstrated by which increased serum uric acid levels could be injurious to vascular endothelium and cardiovascular function. In the present study serum uric acid levels were found to increase profoundly in diabetic subjects compared to non diabetics.

To conclude the serum levels of all the parameters studied in the type 2 diabetic cases were raised except the High density lipoprotein cholesterol when compared to age and sex matched healthy control. Only a group of 120 subjects were studied in the present study and these parameters were found to be statistically significant in the study subjects indicating the role of Lipoprotein(a), Highly sensitive c-reactive protein and Uric acid in addition to lipid profile as cardiovascular risk factors in type2 diabetic subjects.

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