

Assessment of plasma levels of high sensitive c - reactive protein and cholesterol among Sudanese with Type2 diabetes mellitus

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ABSTRACT: A case control study was conducted during the period from March 2012 to June 2012 to assess the plasma levels of high sensitive C-reactive protein and cholesterol among Sudanese with type2 diabetes mellitus. Hundred patients with type2 diabetes mellitus were selected as a test group from the Military Hospital and Zenam Diabetes Center in Khartoum state, Sudan. The test group was compared with a control group which included 50 healthy volunteers. Blood specimens were collected from both groups and fasting blood glucose, glycated hemoglobin, total cholesterol and high sensitive C-reactive protein were estimated. Age and gender of the test group were matched with the control group. Spectrophotometric methods were used for measurement of glucose and total cholesterol. glycated hemoglobin was measured by using NycoCard II device and the high sensitive C-reactive protein was measured by using i-CHROMA device. Statistical package for social science (SPSS version 11.5) computer software was used for data analysis. The results indicated a significant increase in the mean of the plasma levels of high sensitive C-reactive protein of the test group when compared with the control group (3.10 ± 2.69 mg/L) versus (0.47 ± 0.12 mg/L) ($p=0.000$), and a significant elevation in the mean of plasma levels of high sensitive C-reactive protein in diabetic patients with hypertension when compared with the mean of those without hypertension (7.03 ± 1.62 mg/L) versus (1.65 ± 1.06 mg/L) ($p=0.000$). Also there was a significant elevation of the plasma levels of high sensitive C-reactive protein in females with type2 diabetes mellitus when compared with the mean of diabetic males (3.65 ± 2.92 mg/L) versus (2.53 ± 2.34 mg/L) ($p=0.037$). The results indicated a significant, strong positive correlation between high sensitive C-reactive protein in type2 diabetic patients and fasting blood glucose ($r=0.91$, $p=0.000$), glycated hemoglobin ($r=0.88$, $p=0.000$), body mass index (0.91 , $p=0.000$) and the total cholesterol ($r=0.78$, $p=0.000$).

KEYWORDS: *Hyperglycemia, glycated hemoglobin, body mass index.*

INTRODUCTION

Diabetes is a metabolic disorder with inappropriate hyperglycemia either due to an absolute or relative deficiency of insulin secretion or reduction in the biologic effectiveness of insulin or both. It is also associated with disturbances concerned with protein, carbohydrate and lipid metabolism. The decreased uptake of glucose into muscle and adipose tissue leads to chronic extra cellular hyperglycemia which results in tissue damage and chronic vascular

complications in both type I and II Diabetes Mellitus^(1, 2).

Among several markers of inflammation, hs-CRP is found to be significant in people with diabetes. CRP, a pentameric protein produced by the liver has emerged as the golden marker for inflammation'. It is a non-immunoglobulin protein having five identical sub units. It is a member of pentraxin family proteins. The C-reactive protein derives from the fact that it reacts with capsule

polysaccharide of *pneumoniae Streptococcus*. It is an acute phase response protein markedly increased in both inflammatory and infectious diseases. It plays an important role in innate immunity. It assists in complement binding to foreign and damaged cells and enhances phagocytosis⁽³⁾. People with Type 2 diabetes, regardless of blood sugar control, tend to have increased triglycerides, decreased HDL, and increased LDL. The cholesterol profile tends to persist even if blood sugar levels are under control pointing to an even higher likelihood of developing plaques. These LDL molecules are oxidized and taken up by macrophages, which become engorged and form foam cells. These cells often become trapped in the walls of blood vessels and contribute to atherosclerotic-plaque formation. In fact, plaques formed in the arteries of people with Type 2 diabetes tend to be more fatty and less fibrous than in people with Type 1 diabetes, leading to an even higher risk of a plaque dislodging to cause a heart attack or stroke⁽⁴⁾.

The choice was to focus on the plasma levels of high sensitive C-reactive protein and cholesterol among Sudanese with Type2 Diabetes Mellitus for a lot of reasons including, i) rates of diabetes have increased markedly over the last 50 years in parallel with obesity. As on 2011 there are approximately 285 million people with the disease compared to around 30 million in 1985. Long-term complications from high blood sugar can include heart disease, strokes, diabetic retinopathy where eyesight is affected, kidney failure which may require dialysis, and poor circulation of limbs leading to amputations. Type2 diabetes makes up about 90% of cases of diabetes with the other 10% due primarily to diabetes mellitus type 1 and gestational diabetes, ii) CRP level is an independent risk factor for atherosclerotic disease. Patients with high

CRP concentrations are more likely to develop stroke, myocardial infarction, and severe peripheral vascular disease and iii) total cholesterol is important and necessary for human health but high levels of cholesterol in the blood have been linked to damage to arteries and cardiovascular disease.

The objectives of this research were to assess the plasma levels of high sensitive C-reactive protein and cholesterol among Sudanese with Type2 Diabetes Mellitus , measure the plasma level of hs-CRP, HbA1c, FBS, and total cholesterol in Sudanese with Type 2 Diabetes Mellitus , compare the plasma levels of high sensitive C-reactive protein between the test group and the control group , assess the relationship between the plasma level of hs-CRP and body mass index (BMI), total cholesterol, HbA1c, and fasting blood glucose in Sudanese with Type 2 Diabetes Mellitus, assess the association between the hs-CRP and hypertension in Sudanese with Type 2 Diabetes Mellitus and finally determine and compare the hs-CRP between male and female in the test group.

MATERIALS and METHODS

This is a quantitative, descriptive, analytic, case-control and hospital-based study. It was conducted in the military hospital and Zenam Diabetes Centre, located in Khartoum State, during the period March 2012 to May 2012. A total of 100 patients with type 2 diabetes (Test group) were enrolled in this study regularly visit the Military Hospital and Zenam Diabetes Centre for routine follow up. A 50 healthy volunteers, age and sex matched, were included as a control for comparison. Patients with type 1 diabetes mellitus and those with any inflammatory disorders were excluded from this study.

The study was approved by the research board of the Faculty of Medical Laboratory, Sudan University of Science and Technology, and full permission was obtained from the Military Hospital and Zenam Diabetes Centre, Khartoum, Sudan. All participants provided oral consent, for each participant an interview with a questionnaire was used to obtain the clinical data.

Venous blood samples (4mLs) were taken from each participant by standard procedures, divided into two containers, 2mLs in fluoride oxalate anticoagulant container for plasma glucose, and 2mLs in EDTA container for HbA1c (whole blood). The EDTA containers were centrifuged at 3000 rpm for 3minutes; the plasma obtained for CRP and cholesterol, were stored in plain containers which were kept at -80 °C until used.

Spectrophotometric methods were used for measuring glucose and total cholesterol. HbA_{1c} was measured by using chromatographic-spectrophotometric ionexchange method and the hs-CRP was measured by using fluorescence immunoassay technology (sandwich immunodetection method)⁽⁵⁾.

The precision and accuracy of all methods used in this study were checked each time; a

batch was analyzed by including commercially prepared control sera. Statistical Package for Social Science (SPSS version 11.5) computer software was used for data analysis. (Significance levels was set at P≤0.05). Independent t-test, Pearson’s correlation and linear regression were used to compare between means and to assess the correlation between different variables.

RESULTS

This study was conducted on 100 patients with type 2 diabetes as a test group and 50 healthy volunteers as a control group. Age and gender of the test group were matched with control group.

In this study the test group was composed of 49 males (49%) and 51 females (51%), where as the control group was composed of 24 males (48%) and 26 females (52%). Twenty seven percent (n=27) of the patients were hypertensive while 47% (n=47) of them were described as obese according to BMI calculation.

There was a significant difference between the means of high sensitive C - reactive protein in the test group and the control group. (3.10 ±2.69mg/L) versus (0.47± 0.12 mg/L) (p=0.000) (Table 1).

The data also showed a significant

Table 1. Plasma high sensitive C-reactive protein (mg/L) in test and control groups

Variable	Test group (n=100)	Control group (n=50)	P-value
High sensitive C-reactive protein (mg/L)	3.10 ±2.69	0.47± 0.12	0.000

mean ± Standard deviation , P-value ≡ probability value.

difference between the means of high sensitive C-reactive protein of the diabetic patient with hypertension when compared

with those without hypertension (7.03± 1.62 mg/L) versus (1.65± 1.06 mg/L) (p=0.000) (Table 2).

Table 2. Plasma high sensitive C-reactive protein (mg/L) in case groups with and without hypertension

Variable	With hypertension (n=27)	without hypertension (n=73)	P-value
High sensitive C-reactive protein (mg/L)	7.03± 1.62	1.65± 1.06	0.000

mean ± Standard deviation , P-value ≡ probability value.

Differences between the means of high sensitive C-reactive protein in the male (2.53±2.34mg/L) versus female were also significant. (3.65±2.92mg/L) (p=0.037) (Table 3).

Table 3. Plasma high sensitive C-reactive protein (mg/L) according to sex

Variable	Male (n=49)	Female(n=51)	P-value
High sensitive C-reactive protein (mg/L)	2.53±2.34mg/L	3.65±2.92mg/L	p=0.037

mean ± Standard deviation , P-value ≡ probability value.

A significant strong positive correlations between hs-CRP, total cholesterol (r= 0.78, P=0.000) (Fig.1), fasting blood sugar (r=0.91, p= 0.000) (Fig.2), HbA1c (r=0.88, p=0.000)(Figure 3) and body mass index (r=0.91, p=0.000) (Fig 4).

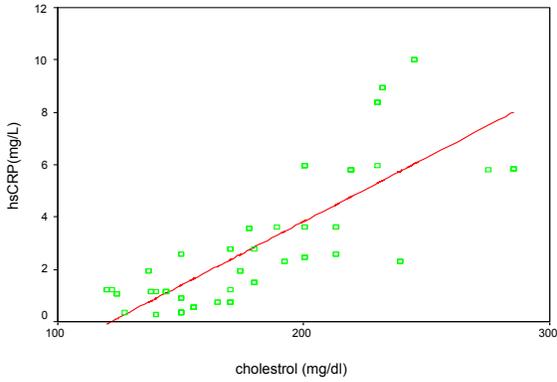


Fig.1. A scatter plot showing the relationship between hs-CRP (mg/L) and total cholesterol (mg/dl) ($r= 0.78$, $P=0.000$).

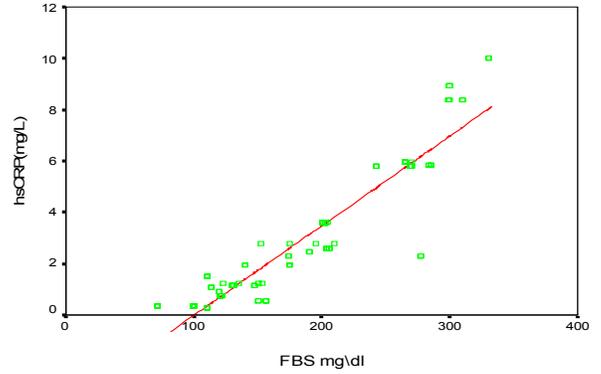


Fig.2. A scatter plot showing the relationship between hs-CRP (mg/L) and fasting blood sugar (mg/dl) ($r=0.91$, $p=0.000$).

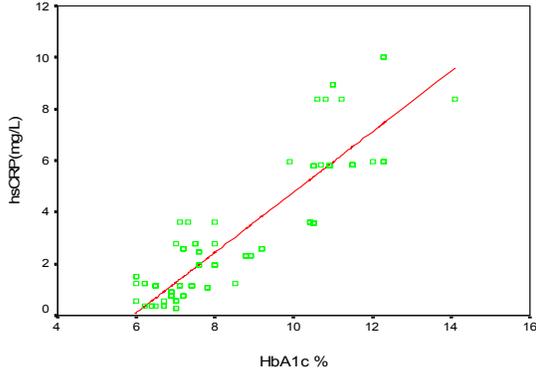


Fig.3. A scatter plot showing the relationship between hs-CRP (mg/L) and HbA1c (%) ($r=0.88$, $p=0.000$)

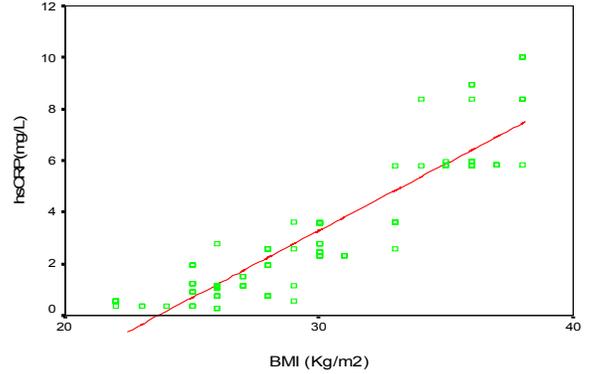


Fig.4. A scatter plot showing the relationship between hs-CRP (mg/L) and body mass index (Kg/m²) ($r=0.91$, $p=0.000$)

DISCUSSION

Diabetes mellitus type 2 (formally non-insulin-dependent diabetes mellitus (NIDDM) or adult-onset diabetes) is characterized by hyperglycemia as a result of an individual's resistance to insulin with an insulin secretory defect. This resistance results in a relative, not an absolute, insulin deficiency. Type2 constitutes the majority of the diabetes cases. However, these patients are more likely to go into a hyperosmolar coma and are at an increased risk of developing macrovascular and microvascular complications⁽⁶⁾.

The elevated cardiovascular risk of diabetic patients is only partially explained by the presence of conventional cardiovascular risk factors, such as glycemic control, lipid abnormalities, hypertension and visceral obesity. This has suggested that additional risk factors, such as genetic risk factors, may favour the increased cardiovascular morbidity and mortality observed in diabetic patients⁽⁷⁾.

In this study patients with type2 diabetes mellitus have a significant increase in the mean of plasma levels of hs-CRP compared with the control subjects ($p=0.000$). This agrees with a study done by Pradhan, *et al.* 2001⁽⁸⁾ who reported that there was a significant elevation of the mean of the hs-CRP in diabetic patients compared to the non-diabetic control ($P=0.001$).

Also there was a significant elevation of the plasma levels of hs-CRP in diabetic patient with hypertension when compared with the mean of those without hypertension ($p=0.000$). This agrees with a study done by Li, *et al.* 2004; Francisco, *et al.*⁽⁹⁾ Taniguchi, *et al.*⁽¹¹⁾ Wu, *et al.*⁽¹²⁾. This due to increase of LDL cholesterol in blood vessels of hypertensive patients which induced the macrophages to recreate

IL6 that induced the liver to recreate C-reactive protein. Also there was a significant elevation of the plasma levels of

hs-CRP in females with type2 diabetes mellitus when compared with the mean of males. ($p=0.037$). This agrees with a study done by Khera, *et al.* 2005⁽¹³⁾. In the current study there was a significant, strong positive correlation between hs-CRP in type2

diabetic patient and fasting blood glucose ($r=0.91, p=0.000$). This finding is consistent with the result obtained by Aronson, *et al.*⁽¹⁴⁾. Furthermore, there was a significant, strong positive correlation between hs-CRP in type2 diabetic patient and HbA1c ($r=0.88, p=0.000$). This agrees with a study done by Li, *et al.*⁽⁸⁾. hs-CRP in type2 diabetic patients was strongly and positively correlated with body mass index ($r=0.91, p=0.000$). The strong positive correlation of hs-CRP with the body mass index is in conformity with the results reported by Mhurchu, *et al.* 2006⁽¹⁵⁾; Eric and John, 2006⁽¹⁶⁾. In the current study there was a significant, strong positive correlation between hs-CRP in type2 diabetic patient and the total cholesterol ($r= 0.78, P=0.000$). This is in line with that reported by Pick, *et al.*⁽¹⁷⁾.

CONCLUSIONS

The hs-CRP levels were invariably higher in diabetic patients and were consistently higher in diabetic patients with hypertension than in those without hypertension. Furthermore, hs-CRP levels in diabetic females were consistently higher than in diabetic males. Moreover, there were strong positive correlation between hs-CRP and fasting blood glucose, HbA1c, body mass index and total cholesterol.

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