ORIGINAL ARTICLE

Serum C-Reactive Protein in Nigerians With Type 2 Diabetes Mellitus

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Abstract

Background: *C-reactive protein is an acute-phase proteins, produce in the liver, its release is stimulated by cytokines (interleukin 6 and tumour necrosis factor alpha). Elevated level of it is a risk factor for coronary heart disease. Baseline levels of C-reactive protein in apparently healthy men and women predict long-term risk of a first myocardial infarction. Diabetics are at increased risk for coronary heart disease, data from the Framingham Study showed a two-to three-fold elevation in the risk of clinically evident atherosclerotic disease in patients with type II diabetes compared to those without diabetes. However, but data regarding CRP in Nigerian diabetic is lacking.*

Method: *A cross-sectional study conducted among patients attending out patient clinic of the Obafemi Awolowo University Teaching Hospitals complex (OAUTHC) Ile Ife, Osun State south western Nigeria. Measurement of C-reactive protein was based on the principle of solid phase enzyme-linked immunosorbent assay (ELISA).*

Results: *A total of 125 consecutive subjects were recruited comprising 75 patients with type II diabetes mellitus with or without hypertension and 50 apparently healthy age-and-sex comparable controls. There was a significant difference between the mean systolic and diastolic blood pressures of the patients and controls. The fasting blood glucose and C-reactive protein were significantly higher in diabetics compared to controls. There was a positive and significant correlation between FBG and CRP in both patients and controls.*

Conclusion: *This study showed that diabetics have significantly higher serum C-reactive protein compared to the apparently controls. Also there was a positive and significant correlation between C-reactive protein and fasting blood glucose among both patients and controls.*

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Introduction

C- reactive protein was first identified by Tilet and Francis in1930 in the plasma of patients with pneumonia and was named for its ability to bind and precipitate the capsular polysaccharide of pneumococcus.[1] It is synthesized in the liver and is normally present as a trace constituent of serum or plasma at levels less than 0.25-1.5µg/ml.[2] Studies have shown that elevated levels of CRP is a risk factor for coronary heart disease (CHD).[4-6] Baseline levels of CRP in apparently healthy men and women predict long-term risk of a first myocardial infarction.[5] In older men and women, elevated CRP was found to be associated with a 10-year risk of CHD regardless of the presence or absence of cardiac risk factors.[6] A single CRP measurement provides information beyond conventional risk assessment, especially in the intermediate Framingham risk men and high Framingham risk women.[6] Elevated CRP levels have also been linked to an increased risk of later development of diabetes mellitus.[7]

Diabetics are at increased risk for CHD, data from the Framingham Study showed a two-to three-fold elevation in the risk of clinically evident atherosclerotic disease in patients with type II diabetes compared to those without diabetes.[8] Diabetic men in the Multiple Risk Factor Intervention Trial (MRFIT) study had an absolute risk of CHD death more than three times higher than that in the non-diabetic cohort, even after adjustment for established risk factors.[9] National cholesterol education program considers diabetes mellitus to be a CHD equivalent in their lipid guideline.[10] Diabetic patients with no history of CHD have the same risk for future myocardial infarction as do non-diabetic patients with known CHD.[11] Diabetes eliminates the usual female advantage in the risk of death from CHD, as these patients have a 5-8 fold higher death rate than do non-diabetic women.[12] Several proposed pathophysiologic mechanisms are implicated in the pathogenesis of vascular disease in diabetes mellitus. These include endothelial dysfunction, diabetic

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dyslipidaemia, hypercoagulability, impaired fibrinolysis, platelet hyperaggregability, oxidative stress and toxic effects of hyperglycaemia.[13]

However study regarding C-reactive protein in Nigerian diabetics is lacking hence decision to carry out this study to find out the relationship between C-reactive and diabetes mellitus in our population.

Materials and Methods

The study design was cross-sectional conducted among patients attending out patient clinic of the Obafemi Awolowo University Teaching Hospitals complex (OAUTHC) Ile Ife, Osun State south western Nigeria. It comprised 75 consecutive patients with type II diabetes mellitus with or without hypertension and 50 apparently healthy age- and sex- comparable controls from the hospital staff and patient relatives who are themselves not relatives of the study patients were recruited. Using a structured pre-evaluated questionnaire, the demographic data, history of cigarette smoking, alcohol consumption, duration of diabetes, duration of hypertension were explored.

The diagnosis of diabetes mellitus was made on the basis of the reported history and medical records. Diabetic with chronic kidney disease, chronic liver disease, congestive cardiac failure or systemic infection were excluded from the study. Also excluded from the study were diabetics on oral contraceptive pills, analgesics or anti-inflammatory drugs and those on HMGcoA reductase inhibitor (statins). Diabetics aged less than eighteen years and those that did not consent were also excluded from the study. Ethical clearance was obtained from the ethics and research Committee of the Obafemi Awolowo University Teaching Hospitals Complex, and all participating subjects signed the informed consent form after being clearly explained to them. The following investigations were carried out: Fasting blood glucose and 2-hour post prandial, fasting lipid, serum electrolytes, urea and creatinine. Urinalysis was done using dip-stick while measurement of C-reactive protein was based on the principle of solid phase enzyme-linked immunosorbent assay (ELISA).

Data Analysis

Data was presented as mean standard deviation (SD). Student t-test was used to test the significance of differences between mean values of continuous variables and Spearman's correlation coefficient was performed to determine the association between variables. With statistical significance set at p (probability) value less than 0.05. Tables and figures were used to present data, Statistical Package for Social Sciences version 11.0 (SPSS Chicago Ill. USA) was used for all statistical analysis.

Results

Demographic and clinical characteristics of the study population

A total of 125 consecutive subjects were recruited comprising 75 patients with type II diabetes mellitus with or without hypertension and 50 apparently healthy age-and-sex comparable controls. Forty-five (60.0%) patients and 31 (62.0%) controls were females with mean ages ± SD of 57.2 ± 9.4 years and 56.6 ± 7.8 years, respectively (p = 0.804). Thirty (40.0%) patients and 19 (38.0%) controls were male with mean ages of 58.3 ± 10.3 years and 58.3 ± 7.3 years, respectively (p = 0.995).

Body mass index differed significantly between patients and controls. The mean BMI of the patients and controls were 26.0±5.1 kg/m^2 and 21.9±1.6 kg/m^2, respectively (p = 0.000). Thirty (40.0%) patients and 48 (96.0%) controls had normal BMI (18.5-24.9 kg/m^2) (Fishers exact test, p = 0.000), 27 (36.0%) patients and 2 (4.0%) controls were overweight (BMI = 25-29.9 kg/m^2) (Fishers exact test, p = 0.000); while, 12 (15.0%) patients were obese (BMI = 30 kg/m^2) and the remaining 6 (8.0%) were underweight (BMI < 18.5kg/m^2). The mean waist circumference of the female patients and controls were 92.5 ± 10.0 cm and 81.5 ± 2.7 cm, respectively (p = 0.000). While male patients and controls had a mean waist circumference of 95.3 ± 7.2 cm and 92.8 ± 2.4 cm, respectively (p = 0.162).

Fifty-two (69.3%) patients were hypertensive-diabetic and 23 (30.7%) were normotensive-diabetic. Thirty-four (65.38%) out of the 52 hypertensive-diabetic were females, while the remaining 18 (34.61%) were males, while 50 (100%) controls were apparently healthy subjects. The mean systolic blood pressure of the patients and controls were 144.0 ± 12.2 mmHg and 120.2 ± 9.1 mmHg, respectively (p = 0.000),while the mean diastolic blood pressure were 87.1 ± 8.0 mmHg and 79.8 ± 8.2 mmHg, respectively (p = 0.000).

Laboratory parameters of the study population

The mean fasting blood glucose of the patients was 9.3

± 2.4 mmol/L and was significantly higher than that of the controls 4.5 ± 1.0 mmol/L (p = 0.000). Patients had significantly higher than controls 2.5 ± 0.5 µg/mL and 1.5 ± 0.4 µg/mL, respectively (p = 0.000). The mean serum total cholesterol of the patients and controls were 5.7 ± 1.3 mmol/L and 3.9 ± 1.2 mmol/L, respectively (p = 0.000). While the serum LDL cholesterol of the patients and controls were 4.0 ± 0.7 mmol/L and 2.1 ± 0.4 mmol/L, respectively (p = 0.000). There was a significant difference in the mean serum triglycerides between patients and controls 2.3 ± 0.5 mmol/L and 1.4 ± 0.2 mmol/L, respectively.(p = 0.000). Serum HDL cholesterol was significantly lower in patients compared to controls 0.9 ± 0.2 mmol/L and 1.78 ± 0.2 mmol/L, respectively. (p = 0.000).

There was a positive and significant correlation between FBG and CRP in both patients and controls (r = 0.656, p = 0.000) and (r = 0.551, (p= 0.000) respectively. Similar correlations were also observed between CRP and BMI, CRP and systolic blood pressure, CRP and diastolic blood pressure in both patients and controls (r = 0.942, p = 0.000) and (r = 0.893, p = 0.000), (r = 0.667, p = 0.000) and (r = 0.738, p = 0.000), (r = 0.438, p = 0.000) and (r = 0.686, p = 0.000), respectively. However, there was no significant correlation between the durations of hypertension and diabetes with CRP among the study patients (r = 0.135, p = 0.251) and (r = 0.039, p = 0.739) respectively.

On regression analysis, BMI, systolic blood pressure, diastolic blood pressure and FBG were significantly associated with CRP among patients (beta value 0.642, p = 0.000), (beta value 0.409, p = 0.001), (beta = 0.162, p = 0.032) and (beta = 0.119, p = 0.036), respectively. Similarly, in the control group, BMI, systolic blood pressure, diastolic blood pressure, and FBG were significantly associated with CRP (beta = 0.765, p =

Table I. Demographic and social characteristics of the study population.

Parameters	Patients	Controls	P-Value
Age			
Male	58.3±10.3	58.3±7.3	0.995
Female	57.2±9.4	56.6±7.8	0.804
Sex			
Male	30(40.0%)	19(38.0%)	0.822
Female	45(60.0%)	31(62.0%)	0.822
BMI (kg/m^2)	26.0±5.1	21.9±1.6	0.000*
<18.5	6 (8.0%)	0 (0.0%)	0.080
18.5-24.9	30 (40.0%)	48 (96.0%)	0.000*
25-29.9	27 (36.0%)	2 (4.0%)	0.000*
30-34.9	8 (10.7%)	0 (0.0%)	0.021*
35-39.9	3 (4.0%)	0 (0.0%)	0.274
>40	1 (1.3%)	0 (0.0%)	1.000
Waist circum-ference (cm)			
Male	95.3±7.2	92.8±2.48	0.162
Female	92.5±1.0	81.5±2.7	0.000*
SBP (mmHg)	144.0±12.2	120.2±9.1	0.000*
DBP (mmHg)	87.1±8.0	79.8±8.2	0.000*

BMI = Body Mass Index, DBP = Diastolic Blood Pressure, SBP = Systolic Blood Pressure

Table II Laboratory parameters of the study population.

Parameters	Patients	Controls	p-value
FBG (mmol/L)	9.3±2.4	4.5±1.0	0.000*
CRP (µg/mL)	2.5±0.5	1.5±0.4	0.000*
Total cholesterol (mmol/L)	5.7±1.3	3.9±1.2	0.000*
LDL cholesterol (mmol/L	4.0±0.7	2.1±0.4	0.000*
HDL cholesterol (mmol/L)	0.9±0.2	1.8±0.2	0.000*
Triglycerides (Mmol/L)	2.3±0.5	1.4±0.2	0.000*
Serum sodium (mmol/L)	134.6±3.2	137.1±3.5	0.000*
Serum potassium (mmol/L)	3.9±4.2	4.1±4.9	0.793
Serum bicarbonate (mmol/L)	22.8±2.7	24.5±2.4	0.000*
Serum urea (mmol/L)	5.3±6.6	3.6±0.6	0.075
Serum creatinine	90.6±37.5	58.2±8.5	0.000*

FBG = Fasting Blood Glucose, CRP = C - reactive protein, LDL = Low Density Lipoprotein, HDL = High Density Lipoprotein, GFR = Glomerular Filtration Rate, * = Significant at p < 0.05

Table II Comparison of CRP levels between hypertensive-diabetic and normotensive-diabetic

Parameter	Hypertensive-diabetic N=52	Normotensive-diabetic N=23	P-value
CRP	2.51±0.53	2.54±0.44	0.823

CRP = C - reactive protein

Table IV Correlation between CRP and systolic blood pressure, diastolic blood pressure, body mass index and fasting blood glucose in the study patients.

Parameters	Spearman correlation Coefficient (r)	P-value
SBP (mmHg)	0.667	0.000*
DBP (mmHg)	0.438	0.000*
BMI (kg/m^2)	0.942	0.000*
FBG (mmol/L)	0.656	0.000*

BMI = Body Mass Index, DBP = Diastolic Blood Pressure, SBP = Systolic Blood Pressure, FBG = Fasting Blood Glucose

Table V Correlation between CRP and systolic blood pressure, diastolic blood pressure body mass index and fasting blood glucose among controls.

Parameters	Spearman correlation coefficient (r)	P-value
SBP (mmHg)	0.738	0.000*
DBP (mmHg)	0.686	0.000*
BMI (kg/m^2)	0.893	0.000*
FBG (mmol/L)	0.551	0.000*

BMI = Body Mass Index, DBP = Diastolic Blood Pressure, SBP = Systolic Blood Pressure, FBG = Fasting Blood Glucose

Table VI Correlation between CRP and duration of hypertension and diabetes in the study patients

Parameters	Spearman correlation coefficient (r)	P Value
Duration of hypertension	0.135	0.251
Duration of diabetes	0.039	0.739

Table VII Multiple regression analysis between CRP and body mass index, systolic blood pressure, diastolic blood pressure and fasting blood glucose among study patients.

Parameters	Beta value	P-value
BMI (kg/m^2)	0.642	0.000*
SBP (mmHg)	0.409	0.000*
DBP (mmHg)	0.162	0.032*
FBG (mmol/L)	0.119	0.036*

BMI = Body Mass Index, DBP = Diastolic Blood Pressure, SBP = Systolic Blood Pressure, FBG = Fasting Blood Glucose

Table VIII Multiple regression analysis between CRP and body mass index, systolic blood pressure, diastolic blood pressure and fasting blood glucose among controls.

Parameters	Beta value	P-value
BMI (kg/m^2)	0.602	0.000*
SBP (mmHg)	0.765	0.001*
DBP (mmHg)	0.689	0.001*
FBG (mmol/L)	0.375	0.000*

BMI = Body Mass Index, DBP = Diastolic Blood Pressure, SBP = Systolic Blood Pressure, FBG = Fasting Blood Glucose

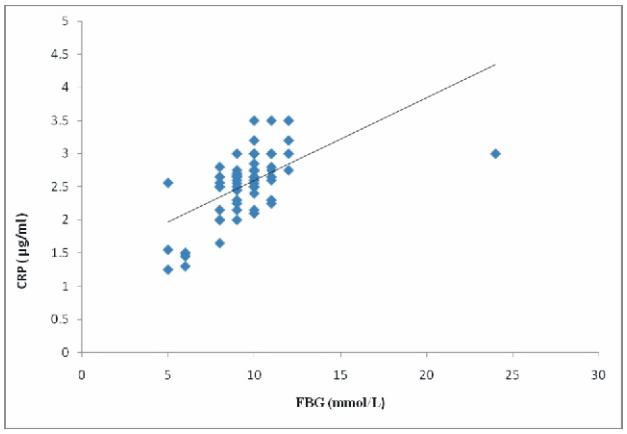

Fig 1 Graphical presentation of correlation between CRP and FBG among patients

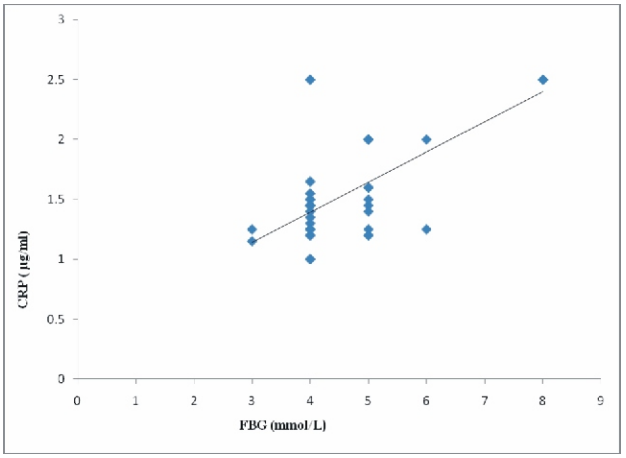

Fig 2 Graphical presentation of correlation between CRP and FBG among controls CRP = C-Reactive Protein, FBG = Fasting Blood Glucose

Discussion

This cross-sectional study of CRP in Nigerians with type II diabetes mellitus with or without hypertension showed that CRP levels are significantly higher in diabetic than controls. Similarly, there was a positive and significant correlation between CRP levels and FBG. Ford[14] had previously reported a similar result, though in his study he used glycated haemoglobin to determine the level of glycaemic control in those with diabetes. This study is limited by lack of facilities to test for glycated haemoglobin. The pathophysiologic mechanisms for the elevated CRP in diabetic is linked to the toxic effects of hyperglycaemia on vascular endothelium, increased oxidative stress and the associated generation of free radicals which is injurious to vascular endothelium and triggers inflammation and cytokines release (interleukin 6, tumour necrosis factor alpha). These cytokines in turn stimulate the synthesis and release of CRP from the liver.[15 and 16]

The study also showed a positive and significant correlation between CRP and systolic, as well as diastolic blood pressure. However, there was no significant difference in the serum CRP levels between hypertensive-diabetic and normotensive-diabetic. This result is consistent with what was previously reported by other workers.[17,18] Similarly, there was no significant correlation between the duration of hypertension and diabetes with CRP among the study patients. This suggests that good glycaemic and blood pressure control are associated with lower serum CRP levels than the absolute duration of hypertension or diabetes. The link between hypertension and CRP was thought to be mediated via angiotensin II. Angiotensin II has, in addition to potent vasoconstricting effect, a proinflammatory effect and CRP has been found to up regulate angiotensin I receptor mRNA and increase the number of angiotensin I receptor binding sites in vascular smooth muscle cells. Angiotensin I receptor is a key atherosclerotic switch facilitating angiotensin-II-induced reactive oxygen species production, vascular smooth muscle cell migration, proliferation, and vascular remodelling.[19]

Lipid abnormalities occur frequently in diabetic, and in this study it was observed that HDL cholesterol level was significantly lower in patients compared to controls. On the other hand, serum total cholesterol, triglycerides and LDL cholesterol levels were significantly higher in patients compared to controls, this result is similar to what was reported by Bruno *et al.*[20] The underlying pathogenesis and the interrelationships between diabetes mellitus and lipid abnormalities have not been

completely elucidated. However, insulin resistance has been hypothesized to be the common underlying pathogenic mechanism.[21]

Conclusion

This study showed that diabetics have significantly higher serum C-reactive protein compared to the apparently controls. Also there was a positive and significant correlation between C-reactive protein and fasting blood glucose among both patients and controls.

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