ORIGINAL ARTICLE

Plasma levels of inflammatory cytokines in adult Nigerians with the metabolic syndrome

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ABSTRACT

Background: The aim of this study is to determine the plasma levels of interleukin 6 (IL-6), tumor necrotic factor alpha (TNF-α), and C-reactive protein (CRP) in adult Nigerians with the metabolic syndrome and to determine the relationship between components of the metabolic syndrome and CRP in adult Nigerians. **Subjects and Methods:** This was a case–control study of fifty adult men and women with the metabolic syndrome, and fifty age- and sex-matched males and females without the metabolic syndrome. Metabolic syndrome was defined based on the National Cholesterol Education Programme-Adult Treatment Panel III criteria. Written informed consent was obtained from the participants. Blood pressure and anthropometry measurements were taken and venous blood was collected after an overnight fast. The Ethics Committee of the Lagos University Teaching Hospital, Lagos, Nigeria, approved the study protocol. Comparisons of continuous variables and categorical variables were done using the Student's *t*-test and Chi-square test, respectively. Regression analysis was used to determine the associations between variables. Statistical significance was set at $P < 0.05$. **Results:** The age- and sex-matched males and females with and without the metabolic syndrome did not differ in their sociodemographic characteristics. They however differed in some clinical and laboratory parameters such as diastolic blood pressure ($P = 0.048$), waist circumference ($P = 0.002$), body mass index ($P = 0.012$), waist/hip ratio ($P = 0.023$), high density lipoprotein (HDL) ($P = 0.012$), and insulin resistance (IR) ($P = 0.042$). There was a statistically significant increase in the inflammatory marker, CRP ($P = 0.019$), the cytokines, IL6 ($P = 0.040$), and TNF-α ($P = 0.031$) between the subjects with and without metabolic syndrome. There was also a significant association between CRP, waist circumference, IR, and HDL in the metabolic syndrome ($P < 0.05$). **Conclusion:** Plasma levels of inflammatory cytokines are raised in metabolic syndrome and this may provide novel strategies for the management of metabolic syndrome and related disorders.

Key words: Inflammation, insulin resistance, metabolic syndrome, obesity

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INTRODUCTION

Metabolic syndrome was first described by Reaven[1] in 1988 when he described the clustering of metabolic abnormalities of insulin resistance (IR)/glucose intolerance, hypertension, dyslipidemia (high triglyceride and low high-density lipoprotein [HDL] cholesterol concentrations) and obesity in one individual. The components of the syndrome are risk factors for atherosclerosis, making metabolic syndrome a significant risk for coronary heart disease.[2] Obesity and IR also provide significant risk for developing type 2 diabetes.[3] Reaven called it the IR syndrome because he believed that IR accounted for every component of the syndrome.[1]

Other new features have been added to the metabolic syndrome criteria over time such as increased plasminogen

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activator inhibitor 1 and more recently increased C-reactive protein (CRP) simply because they were frequently found in association with the metabolic syndrome. It is now hypothesized that the presence of inflammation in the metabolic syndrome can explain these new relationships and existing ones.[4]

There has been an ongoing debate about the cause of the onset of the metabolic disturbances that constitute the syndrome, and there have been several attempts to define the metabolic syndrome with special attention to one or another component. The National Cholesterol Education Programme-Adult Treatment Panel III (NCEP-ATPIII) in 2002[5] gave equal weight to each component of the syndrome requiring a combination of at least any 3 of the 5 criteria to make a diagnosis; abdominal circumference ≥102 cm in males or ≥88 cm in females, HDL cholesterol <1.03 mmol/L (<40 mg/dL) (males) or <1.3 mmol/L (<50 mg/dL) (females), triglycerides ≥1.7 mmol/L (≥150 mg/dL), blood pressure ≥130/85 mmHg, or the patient receiving hypotensive treatment and fasting glycaemia >6.1 mmol/L (>110 mg/dL). The International Diabetic Federation in 2005 published their guideline[6] for diagnosis of metabolic syndrome that had race/region specific cut-offs for abdominal obesity, abdominal obesity being the first requirement for diagnosis plus any other two components of the syndrome. The World Health Organization criteria have IR as a mandatory criterion.[7]

Current concepts on the pathophysiology of the metabolic syndrome have it that inflammation is the link between abdominal obesity, IR and cardiovascular disease in the metabolic syndrome.[4] Tumor necrotic factor alpha (TNF-α) and interleukin 6 (IL-6) are proinflammatory cytokines which have been linked with abdominal obesity and the metabolic syndrome.[8-11] IL-6 has been linked with increased production of CRP in the liver, atherosclerosis, and cardiovascular mortality.[12] Current concepts of insulin as an anti-inflammatory hormone have been reported[4,8] and impairment of insulin action in IR occasioned by the proinflammatory state of excess adiposity would explain the link between abdominal obesity, IR, and the metabolic syndrome.[4]

There are few studies on the relationship between inflammation and the metabolic syndrome in adult Nigerians. This study aims to determine the plasma levels of IL-6, TNF-α, and CRP in adult Nigerians with the metabolic syndrome and to determine the relationship between components of the metabolic syndrome and CRP in adult Nigerians. The awareness of the importance of inflammation in the metabolic syndrome may help to develop new strategies for the prevention and treatment of metabolic syndrome and related disorders.

SUBJECTS AND METHODS

This case–control study consisted of fifty adult men and women with the metabolic syndrome and fifty age- and sex-matched males and females without the metabolic syndrome. The diagnosis of the metabolic syndrome was based on the NCEP-ATPIII criteria.[5] The subjects with metabolic syndrome were drawn from patients attending the Obesity and Metabolic Clinic of the Lagos University Teaching Hospital and the controls were members of staff of the hospital. The Ethical Research and Review Committee of the Hospital approved the study protocol, and informed consent was obtained from the participants.

The inclusion criterion was adult males and females between 30 and 70 years of age. Known diabetics were excluded from the study. The study participants reported on the morning of the study after an overnight (10–12 h) fast. Five milliliters of venous blood was collected from the antecubital vein.

Abdominal obesity was determined by measurement of the waist circumference in centimeters using the pubic crests and the umbilicus as landmarks.The hip circumference was measured from the farthest point on the gluteus using the anatomical neck of the femur as landmarks. The blood pressure was determined using the Accoson's Mercury Sphygmomanometer (cuff size 15 cm × 43 cm). The subjects were seated and rested for 30 min before measurement. The systolic blood pressure was taken at the first korotkoff sound and diastolic at the fifth korotkoff sound. The average of two readings taken 15 min apart was used.

The total, low-density lipoprotein (LDL), HDL cholesterol, triglyceride, and glucose concentrations were determined on fasting serum samples using reagents from Randox Laboratories Limited, Antrim, UK, BT 29 4QY, on semiautomatic biochemistry analyzer BS3000P-Sinnowa Medical Science and Technology Company Limited, Nanjing, China (211135). Serum levels of IL-6, TNF-α, CRP, and insulin were determined using reagents from Biovendor Laboratories, 62100 Brno, Czech Republic by an enzyme-linked immunoassay technique[13] on Acurex Plate Read - Acurex Diagnostics, Ohio, USA (419-872–4775). IR was calculated using the homeostasis model assessment for IR formula: (Fasting glucose [mmol/L]) × (fasting insulin [μU/mL])/22.5.

The data were analyzed using thestatistical package for the Social Sciences Software (version 20.0; SPSS Inc., Chicago, IL) package. Independent Student's *t*-test was used to test the differences in the mean values for the continuous variables. Chi-square test was used to test the differences in the proportion of the categorical variables. Regression analysis was used to determine the association between variables. Statistical significance was set at $P < 0.05$.

RESULTS

The study population included twenty men and thirty women with metabolic syndrome mean age of 47.84 ± 6.4 years and age and sex matched controls. Table 1 shows the sociodemographic characteristics of the study participants.

The age- and sex-matched cases and controls did not differ in their sociodemographic characteristics.

Table 2 shows the clinical and laboratory characteristics of the study participants.

The age- and sex-matched cases and controls differed in some of the metabolic syndrome parameters. The inflammatory markers, IL-6, TNF-α and CRP, were significantly higher in the group with metabolic syndrome.

Table 3 shows the regression of CRP on components of the metabolic syndrome.

There was an association between CRP and waist circumference, HDL and IR.

Table 1: Sociodemographic characteristics of the study participants

Characteristics	Metabolic syndrome *n*=50 (%)	No metabolic syndrome *n*=50 (%)	*P*
Sex			
Females	30 (60)	30 (60)	1.0
Males	20 (40)	20 (40)	
Ethnicity			
Yoruba	29 (58)	34 (68)	0.567
Igbo	15 (30)	11 (22)	
Others	6 (12)	5 (10)	
Type of work			
Sedentary	40 (80)	39 (78)	0.806
Nonsedentary	10 (20)	11 (22)	
Level of education			
Primary	0 (0)	1 (2)	0.60
Secondary	17 (34)	15 (30)	
Polytechnic/university	33 (66)	34 (68)	
Religion			
Christianity	43 (86)	39 (78)	0.297
Islam	7 (14)	11 (22)	
Marital status			
Married	44 (88)	39 (78)	0.183
Single	6 (12)	11 (22)	
Smoking			
No	45 (90)	49 (98)	0.159
Stopped	2 (4)	1 (2)	
Yes	3 (6)	0 (0)	
Alcohol			
No	34 (68)	36 (72)	0.082
Occasional	5 (10)	10 (20)	
Yes	11 (22)	4 (8)	

Table 2: Clinical and laboratory characteristics of the study participants

Characteristics	Mean±SD		*P*
	Metabolic syndrome (*n*=50)	No metabolic syndrome (*n*=50)	
Age (years)	48.32±6.62	47.84±6.43	0.80
SBP (mmHg)	131.60±16.36	124.52±19.7	0.18
DBP (mmHg)	83.42±11.49	76±13.56	0.048*
WC (cm)	104.73±12.94	99.10±0.07	0.002*
BMI (kg/m2)	30.59±4.64	24.57±10.92	0.012*
WHR	0.88±0.04	0.84±0.05	0.023*
Glucose (mmol/L)	4.88±1.2	4.75±2.79	0.82
TG (mmol/L)	1.92±0.12	1.83±0.21	0.10
HDL (mmol/L)	1.26±0.12	1.36±0.13	0.012*
TC (mmol/L)	5.08±0.46	5.2±0.40	0.36
LDL (mmol/L)	2.94±0.44	2.99±0.43	0.66
IR	5.44±1.19	3.53±1.27	0.042*
CRP (mg/L)	6.59±5.71	3.11±1.82	0.019*
IL-6 (pg/ml)	11.94±2.50	5.13±2.8	0.04*
TNF-α (pg/ml)	8.30±3.26	4.15±2.11	0.031*

*Statistically significant. IR – Insulin resistance; SBP – Systolic blood pressure; DBP – Diastolic blood pressure; HDL – High density lipoprotein; LDL – Low density lipoprotein; BMI – Body mass index; WHR – Waist/hip ratio; TC – Total cholesterol; TG – Triglyceride; CRP – C-reactive protein; WC – Waist circumference; SD – Standard deviation; TNF-α – Tumor necrotic factor-alpha

DISCUSSION

This study reports significantly elevated levels of the proinflammatory cytokines IL-6, TNF-α and the acute phase protein, CRP in the subjects with metabolic syndrome compared to the control subjects without metabolic syndrome. This is similar to the findings by Indulekha *et al.*[14] and Choi *et al.*[15] reported significantly elevated high-sensitivity CRP levels in elderly Korean women with impaired glucose tolerance compared to controls with normal glucose tolerance but also reported comparable levels of TNF-α and IL-6 in women with and without impaired glucose tolerance. Kitsios *et al.*[16] carried out a study on obese young adults and reported elevated IL-6 levels but comparable TNF-α levels between obese and normal weight young adults. The different study populations, the different criteria for defining metabolic syndrome as endorsed by different organizational bodies,[5-7] and the different combinations of dysmetabolic features that characterize the syndrome may account for some of the observed differences from these studies. A recurring factor from these studies, however, is an increase in the concentration of one or more markers of inflammation in relation to the different components that make up the syndrome.

The reason for the inflammation in the metabolic syndrome is not yet fully understood. An explanation may be that larger adipose tissue mass in obesity leads to increased release of IL-6 and TNF-α into the circulation which in turn accounts for a greater production of CRP by the liver.[4,8] Another possibility is that IR itself is responsible for the higher production of the cytokines.[4,8]

These reports corroborate our findings in this study of a positive association between CRP and waist circumference,

Table 3: Regression of C-reactive protein on components of the metabolic syndrome

Metabolic syndrome components	Regression coefficient	95% CI	*P*
SBP	−0.16	−0.77-0.43	0.57
DBP	−0.10	−0.52-0.31	0.62
WC	0.41	0.20-0.50	0.025*
BMI	0.177	−0.001-0.35	0.051
Glucose	0.05	−0.01-0.14	0.129
TG	−0.002	−0.007-0.003	0.47
HDL	−0.0048	−0.009-−0.007	0.023*
IR	0.084	0.003-0.17	0.048*

*Statistically significant. CI – Confidence interval; SBP – Systolic blood pressure; DBP – Diastolic blood pressure; HDL – High-density lipoprotein; WC – Waist circumference; IR – Insulin resistance; TG – Triglyceride; BMI – Body mass index; WC – Waist circumference

a surrogate marker for abdominal obesity, and IR in the metabolic syndrome.

The original description of the metabolic syndrome by Reaven[1] consisted of a clustering of dysmetabolic features accounted for by resistance to the classic metabolic functions of insulin. Thus, hyperinsulinemia, glucose intolerance, type 2 diabetes, hypertriglyceridemia, and low HDL concentrations can be accounted for by resistance to the actions of insulin on glucose and carbohydrate metabolism.[17] The defects of insulin action in glucose metabolism include failure to suppress gluconeogenesis in the liver and failure to mediate glucose uptake in insulin-sensitive tissues (i.e., muscle and adipose tissue). To compensate for defects in insulin action, insulin secretion must be increased to sustain euglycemia, leading to a state of hyperinsulinemia. Failure of this compensatory mechanism will result in glucose intolerance and hyperglycaemia.[17]

In the adipocytes, insulin enhances the incorporation of free fatty acids into triglycerides by its activation of lipoprotein lipase, insulin also inhibits the activity of hormone sensitive lipase thereby decreasing the efflux of free fatty acids from adipocytes.[18] In a state of IR, the adipocytes are resistant to the effects of insulin. The increased free fatty acid flux to the liver causes increased hepatic very LDL (VLDL) production. A higher proportion of triglyceride is transferred from the triglyceride-rich VLDL to LDL and HDL by the cholesteryl ester transfer protein. The hydrolysis of the triglyceride-rich LDL produces a preponderance of small dense HDL particles that is filtered by the kidney resulting in low HDL concentrations.[18] The increased free fatty acid flux worsens the insulin resistant state through specific actions that block insulin signal transduction.[4]

The finding of increased CRP levels and an association between CRP, a marker of inflammation and the metabolic syndrome components of waist circumference, IR and low HDL, a marker of cardiovascular risk, in this study, supports reports from other studies of the inclusion of elevated levels of CRP as a new feature associated with the metabolic syndrome.[4,8]

Current concept of insulin as an anti-inflammatory hormone and obesity as a proinflammatory condition provide a conceptual framework with which to place a substantial number of apparently unrelated biological events into a pathophysiological construct and account for the link between inflammation, abdominal obesity, IR, and cardiovascular disease in the metabolic syndrome.[19]

Novel nonmetabolic actions of insulin as an anti-inflammatory hormone have been supported by recent observations that insulin has been shown to suppress several proinflammatory transcription factors and the genes regulated by them,[20,21] an impairment of insulin action in IR would thus lead to the activation of these proinflammatory transcription factors and expression of their corresponding genes. Further studies have also shown that insulin reduced the plasma concentrations of CRP and other inflammatory mediators in subjects with type 2 diabetes and severe hyperglycemia[22,23] and recent observations on the interference of insulin signal transduction by inflammatory mechanisms in obesity further supports the inflammation hypothesis.[24]

Observations made in the USA on patients with the metabolic syndrome, who were being treated for inflammatory arthritis, with the anti-inflammatory drug etanercept, revealed that the patients had reduced levels of CRP and other inflammatory cardiovascular risk markers following weeks of therapy.[25] This underscores the place of inflammation in the metabolic syndrome and its potential for therapy for metabolic syndrome and related disorders.

CONCLUSION

This study reports an increase in inflammatory mediators in the metabolic syndrome. It also shows a statistically significant association between CRP and some components of the metabolic syndrome. Inflammation may have a role to play in the pathogenesis of the disorder.

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Conflicts of interest
There are no conflicts of interest.

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