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SERUM ADIPONECTIN LEVELS IN TYPE 2 DIABETIC PATIENTS WITH HYPERTENSION IN ZARIA, NIGERIA

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ABSTRACT

Background: Hypertension commonly occurs in type 2 diabetes mellitus. Adiponectin, a recently discovered protein that is secreted by adipocytes has been demonstrated to have both antidiabetic and antiatherogenic properties. Recent studies have also described an association between adiponectin and hypertension; though results have been majorly inconsistent. This current study is aimed at evaluating the serum adiponectin levels in type 2 diabetics with hypertension in Zaria, Nigeria. **Materials and Methods:** Serum adiponectin, glycated haemoglobin, fasting plasma glucose and anthropometric parameters were measured in 90 type 2 diabetic and 90 apparently healthy (control) subjects. Adiponectin was measured using the ELISA method while glycated haemoglobin and fasting plasma glucose were measured using spectrophotometric methods. The results were statistically analysed using the two-tailed student's t-test and the one way analysis of variance (ANOVA), and relationships investigated using Pearson's correlation. **Results:** Values of serum adiponectin, glycated haemoglobin, fasting plasma glucose and body mass index (BMI) were significantly higher ($P < 0.05$) in type 2 diabetic subjects than similar values in the apparently healthy controls. Values were similar among the type 2 diabetic hypertensive and their non-hypertensive counterparts ($P>0.05$). Mean ± SEM values of serum adiponectin, glycated haemoglobin, fasting plasma glucose and BMI all showed no significant differences ($P > 0.05$) between the type 2 diabetic hypertensive and the type 2 diabetic non- hypertensive subjects. No significant differences in mean ± SEM values were also found in serum adiponectin, glycated haemoglobin, fasting plasma glucose and BMI across the classification of hypertension ($P > 0.05$), within both the type 2 diabetic hypertensive group and the few non - diabetic hypertensive controls. Pearson's correlation found a significant negative relationships between adiponectin and hypertension ($r = -0.192$) and a significant positive relationship between hypertension and BMI ($r = 0.297$). **Conclusion:** Our results did not reveal any significant differences in mean values of adiponectin between the hypertensive and non-hypertensive type 2 diabetics in our environment. Further study is suggested with a larger sample size.

INTRODUCTION

In order to understand hypertension or high blood pressure, it is very important to understand what blood pressure (BP) is. Blood is carried from the heart to all parts of the body in blood vessels. Each time the heart beats, it pumps blood into the vessels. BP is created by the force of blood pushing against the walls of blood vessels (arteries) as it is pumped by the heart.[1] There are many definitions of hypertension in literature. However, the definitions proposed by the Joint National Committee VII (*2*) and WHO *(1)* are two popular definitions that are in use.

According to WHO (2003), hypertension is defined as a systolic BP equal to or above 140 mmHg and/ or diastolic BP equal to or above 90 mmHg. JNC VII has proposed another definition of hypertension that might be clearer than the definition of WHO because of the classification of BP. According to JNC VII, hypertension is defined as systolic BP level of ≥ 140 mmHg and diastolic BP of ≥ 90 mmHg. The JNC VII defined normal BP as a systolic BP < 120 mmHg and diastolic BP < 80mmHg. The area between systolic BP of 120-139 mmHg and diastolic BP of 80-89 mmHg is defined as "prehypertension".[2]

The World Health Organization's recent fact sheet says that worldwide obesity has nearly tripled since 1975. In 2016, more than 1.9 billion adults, 18 years and older, were overweight. Of these, over 650 million were obese. Thirty nine percent (39%) of adults aged 18 years and over were overweight in 2016, and 13% were obese. Thirty nine (39) million children under the age of 5 were overweight or obese in 2020. Over 340 million children and adolescents aged 5-19 were overweight or obese in 2016. Overweight and obesity are defined as abnormal or excessive fat accumulation that may impair health.[3] Because obese individuals are at risk of diabetes, hypertension, arteriosclerosis and other cardiovascular diseases, obesity and obesity-related diseases are a worldwide public health problem.

Adipose tissue is involved in regulating a variety of homeostatic processes as an endocrine organ that secretes many biologically active molecules. Adiponectin is a recently discovered protein that seems to be exclusively secreted by adipocytes and is the most abundant adipose tissue–derived protein.[4,5] Plasma adiponectin levels in humans have been found to be lower in obese subjects than in their non-obese counterparts[4,6], in patients with coronary artery disease[7,8] and type 2 diabetes mellitus than in healthy subjects.[9 – 11] It has also been found to be higher in women than in men.[12]

Hypertension is a major trigger of cardiovascular complications and is associated with endothelial dysfunction and aterosclerosis. Several cross sectional studies have shown that adiponectin correlates negatively with blood pressure but the results of studies on the relationship between adiponectin and hypertension have remained inconsistent probably due to small-sample clinical studies which are not sufficient to establish a cause-effect relationship.[13,14]

Hypertension is common among patients with diabetes with the prevalence depending on type and duration of diabetes, age, sex, race/ethnicity, BMI and history of glycaemic control, among other factors.[15,16] In this paper, we present the findings of the cross sectional study that we did to investigate the relationship between adiponectin, a key player in type 2 Diabetes Mellitus, and hypertension among type 2 Diabetic subjects in Zaria, Nigeria.

MATERIALS AND METHODS

A total of 180 subjects were recruited for the study. These comprised 90 (21 male, 69 female; age: 25 – 70 years) consecutive type 2 diabetic patients attending the diabetic clinic at the Ahmadu Bello University Teaching Hospital, Zaria and 90 (27 male, 53 female; age: 24 – 65 years) apparently healthy subjects from the Ahmadu Bello University Teaching Hospital, Zaria and its environs as controls. Ethical approval was obtained from the Ethical committee of the Faculty of Medicine (now College of Medical Sciences) ABU/ABUTH, Zaria. Ten (10) microliters (mls) of fasting venous blood was collected from each subject. Seven (7) mls was dispensed into a plain blood sample bottle appropriately labelled for the subject. This was allowed to clot and the serum was harvested after spinning in the centrifuge at 3000 r.p.m for 5 minutes. Three (3) microliters (mls) of blood was dispensed into a fluoride oxalate bottle, which was used for glycated haemoglobin. The plasma was extracted after spinning at 3000 r.p.m for 5 minutes and was used for glucose estimation. Fasting plasma glucose (FPG), glycated haemoglobin (HbA1c), and adiponectin were measured for all subjects in the study.

Full medical history of recruited subjects was obtained including weight, height and blood pressure measurements. Body mass index (BMI) was calculated as the value of weight (kg) divided by height^2 (m^2).

Adiponectin was measured by the ELISA method of Andersen *et al*,[17], as modified by WKEA Med Supplies Corp. Fasting plasma glucose was estimated using the enzymatic glucose oxidase - peroxidase method.[18] Glycated haemoglobin was measured using the Ion Exchange Resin method.[19]

The data obtained were analyzed on Windows Seven using Statistical Package for Social Sciences (SPSS) version 20.0 (2013). The results were compared using the two-tailed Student's t-test and one way analysis of variance (ANOVA). A P- value of equal to or less than 0.05 ($P \leq 0.05$) was considered as statistically significant. A correlation analysis was done to evaluate the relationship between adiponectin and hypertension in both the diabetic and apparently healthy (control) groups using Pearson's correlation.

RESULTS

The results obtained in the present study are presented as tables 1 – 8.

Table 1. Clinical parameters (mean ± SEM) in diabetic patients and apparently healthy (control) group. The mean values of the parameters were significantly higher ($P < 0.05$) in the diabetic patients than similar values in the controls except for age.

Table 2. Mean values ± SEM of FPG, HbA1c and Adiponectin in diabetic subjects and control group. The mean values (± SEM) of FPG and HbA1c are significantly higher ($P < 0.05$) in the diabetic than the non – diabetic subjects. On the other hand, the mean value (± SEM) of adiponectin in the diabetic subjects is significantly lower ($P < 0.05$) than in the control subjects.

Table 3. Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI in hypertensive and non – hypertensive type 2 diabetic subjects.There is no significant difference ($P > 0.05$) in the mean values of the parameters in both the hypertensive and non – hypertensive type 2 diabetic subjects.

Table 4. Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI in hypertensive and non – hypertensive apparently healthy subjects (control group). There is no significant difference ($P > 0.05$) in the mean values of the parameters in both the hypertensive and non-hypertensive control subjects.

Table 5. Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI according to classification of hypertension in type 2 diabetic subjects. There is no significant difference ($P > 0.05$) in the mean values of the parameters across the groups.

Table 6. Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI according to classification of hypertension in the apparently healthy (control) subjects. The mean

values of all the parameters were similar across the classification in the apparently healthy (control) subjects. No significant differences in mean values were observed (P > 0.05).

Table 7. Pearson's correlation showing the relationship between hypertension and adiponectin. Pearson's correlation shows a significant negative relationship between hypertension and adiponectin (r = -0.192).

Table 8. Pearson's correlation showing the relationship between hypertension and BMI. A significant positive relationship is observed between hypertension and BMI (r = 0.297).

DISCUSSION

Decreased serum levels of adiponectin has been documented in obesity, a risk factor for the development of diabetes mellitus, dyslipidaemia, hypertension and atherosclerosis.[20,21] In obese patients, visceral body fat may affect health conditions, through an abnormal production of adipokines. Adiponectin plays a pivotal role in energy metabolism. Concentration of both total adiponectin and HMW chains decreases in obesity and increases after weight loss.[22,23,24] In addition, total and HMW adiponectin oligomers are inversely correlated to BMI, glucose, insulin and triglyceride levels, degree of IR, and, importantly, visceral fat accumulation.[24]

Our work shows a significant difference in adiponectin (P < 0.002), BMI, SBP, DBP, FPG and HbA1c (P < 0.000) between the diabetic subjects and the non-diabetic (apparently healthy) control group, in agreement with works that have been done by other researchers.[5,11,25-30]

Some clinical and experimental studies have suggested that adiponectin may play a pivotal role in the development of vascular and metabolic diseases. An association between hypertension and adiponectin levels has been reported by several groups. Kazumi *et al*. suggested that young Japanese men with high-normal blood pressure had lower adiponectin levels.[31] Furuhashi *et al* reported that adiponectin levels were significantly decreased in young, non-obese, normotensive men with a family history of essential hypertension.[32] Lee *et al*. reported that higher adiponectin levels were independently associated with a lower risk of hypertension.[33] The mechanism by which adiponectin levels are lowered in patients with hypertension remains to be clarified, although there are several mechanisms that could account for the relationship. First, activation of the renin-angiotensin system (RAS) may be induced in adipose tissue by hypoadiponectinaemia, resulting in an increase in fat mass and blood pressure.[34] RAS blockade has been reported to increase adiponectin levels in essential hypertension.[35] Second, some studies have indicated that inflammation contributes to the pathogenesis of hypertension. Since adiponectin seems to have several beneficial and protective effects, including anti-inflammatory, vasculoprotective and anti-diabetic effects, reduced adiponectin levels may well contribute to the development of hypertension. The antiatherogenic properties of adiponectin are mainly due to NO production in endothelial cells, using phosphatidylinositol 3-kinase-dependent pathways, as well as AMPK pathway.[36,37] The NO production, in physiologic condition, relaxes vessels and exerts anti-inflammation and antithrombotic effects on the vascular wall.[38] In addition, adiponectin decreases smooth muscle cell proliferation and TNFα expression in macrophages.[39] Further study is however, required on this.

Our results, however, did not find any significant differences in the mean serum levels of adiponectin, FPG, HbAIc and BMI (P > 0.05) between the hypertensive and non- hypertensive type 2 diabetic subjects. This could be due to alterations in glucose metabolism observed in type 2 diabetes. Fawwad *et al* demonstrated a loss of correlation with clinical parameters of metabolic syndrome in type 2 diabetes.[40] They posited that this could be as a result of alterations in glucose metabolism seen in diabetes or the effect of medications taken for diabetes or even hypertension.

No significant differences were found between the mean values of adiponectin, HbA1c, FPG and BMI (P > 0.05) in hypertensive and non – hypertensive apparently healthy control subjects. Similarly, no significant differences (P > 0.05) were found in the test parameters (adiponectin, HbA1c, FPG and BMI) across the classes of hypertension between the hypertensive and the non – hypertensives in both the diabetic and the apparently healthy control group. This could be due to the small sample size in the present study.

Pearson's correlation, however, shows a significant negative relation between adiponectin and hypertension at the 0.01 level (r = -0.192), and a significant positive relationship between hypertension and BMI (r = 0.297).

Hypertension is often found co-existing with diabetes mellitus. This is as a result of the endothelial damage that often occurs in diabetes mellitus. Adiponectin, by its antiatherogenic and antidiabetic effects can help to ameliorate these two conditions. More research should be done involving larger sample sizes to investigate the role of adiponectin in the development and management of these conditions and to clearly establish the relationship between adiponectin and these conditions.

Table 1: Clinical parameters (mean ± SEM) in diabetic patients and apparently healthy (control) group.

Subjects	n	Age (years)	BMI (Kg/m^2)	DODM (months)	SBP (mmHg)	DBP (mmHg)
Diabetic patients	90	52.07±1.12	29.54±0.65	72.87±8.12	140.83±2.57	89.09±1.50
Controls	90	51.16±0.99	25.48±0.42	-	123.83±1.79	79.20±1.26
P – value		0.061(NS)	0.000(S)		0.000(S)	0.000(S)

n = Number of subjects
BMI = Body Mass Index
DODM= Duration of Diabetes mellitus
SBP= Systolic Blood Pressure
DBP= Diastolic Blood Pressure
SEM= Standard Error of Mean
(P< 0.000) shows significant differences in the clinical parameters between the diabetic and control groups.
S = Statistically Significant (P < 0.05)
NS = Not significant (P > 0.05)

Table 2: Mean values ± SEM of FPG, HbA1c and Adiponectin in diabetic subjects and control group.

Subjects	n	FPG (mmol/L)	HbA1c (%)	Adiponectin (µg/mL)
Diabetics	90	8.92±0.45	7.39±0.21	1.06±0.06
Controls	90	4.78±0.07	5.53±0.17	1.49±0.13
P – value		0.000(S)	0.000(S)	0.002 (S)

n=Number of subjects
FPG= Fasting Plasma Glucose
HbA1c=Glycated Haemoglobin
SEM= Standard Error of Mean
(P < 0.000) for FPG, HBA1c, and (P < 0.002) for Adiponectin are all significant indicating a significant difference in the diabetic patients and the control group.
S = statistically significant (P < 0.05).

Table 3: Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI in hypertensive and non - hypertensive type 2 diabetic subjects.

	n	Adiponectin (µg/ml)	HbA1c (%)	FPG (mmol/l)	BMI (Kg/m^2)
Hypertensives	71	1.04±0.06	7.31±0.24	8.70±0.50	29.40±0.73
Non Hypertensives	19	1.16±0.14	7.71±0.51	9.74±1.03	30.08±1.49
P-Value		0.575(NS)	0.404(NS)	0.600(NS)	0.518(NS)

n = Number of subjects
S.E.M. = Standard Error of Mean
FPG = Fasting Plasma Glucose
HbA1c = Glycated Haemoglobin
BMI = Body Mass Index
P – Value = Shows level of significance in differences between groups
S = Significant (P < 0.05)
NS = Not significant (P > 0.05)

Table 4: Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI in Hypertensive and Non hypertensive apparently healthy subjects (control group).

	n	Adiponectin (µg/ml)	HbA1c (%)	FPG (mmol/l)	BMI (Kg/m^2)
Hypertensives	16	1.31±0.17	5.80±0.46	5.08±0.21	28.79±1.29
Non Hypertensives	74	1.53±0.15	5.48±0.18	4.71±0.07	24.77±0.28
P-Value		0.214(NS)	0.222(NS)	0.315(NS)	0.401(NS)

n = Number of subjects
S.E.M. = Standard Error of Mean
FPG = Fasting Plasma Glucose
HbA1c = Glycated Haemoglobin

BMI = Body Mass Index
P – Value = Shows level of significance in differences between groups
NS = Not significant (P > 0.05)

Table 5: Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI according to classification of hypertension in type 2 diabetic subjects.

Classification of Hypertension	n	Adiponectin (μg/ml)	HbA1c (%)	FPG (mmol/l)	BMI (Kg/m^2)
Normal SBP <120; DBP < 80	12	1.18±0.18	7.47±0.85	8.61±1.15	29.23±1.60
Pre Hypertension SBP 120 – 139; DBP 80 – 89	29	1.01±0.07	7.90±0.39	9.46±0.87	28.04±0.91
Stage 1 SBP 140 -159; DBP 90 – 99	25	1.06±0.09	6.28±0.28	7.05±0.55	31.53±1.69
Stage 2 SBP > 160; DBP > 100	24	1.08±0.16	7.34±0.40	10.36±0.97	29.45±0.96
P – Value		0.851(NS)	0.288(NS)	0.045(NS)	0.230(NS)

n = Number of subjects
S.E.M. = Standard Error of Mean
SBP = Systolic Blood Pressure
DBP = Diastolic Blood Pressure
FPG = Fasting Plasma Glucose
HbA1c = Glycated Haemoglobin
BMI = Body Mass Index
P – Value = Shows level of significance in differences between groups
NS = Not significant (P > 0.05)

Table 6: Mean values ± SEM of Adiponectin, HbA1c, FPG and BMI according to classification of hypertension in the apparently healthy (control) subjects.

Classification of Hypertension	n	Adiponectin (μg/ml)	HbA1c (%)	FPG (mmol/l)	BMI (Kg/m^2)
Normal SBP <120; DBP < 80	4	1.62±0.21	5.52±0.22	4.71±0.09	24.67±0.62
Pre Hypertension SBP 120 – 139; DBP 80 – 89	24	1.23±0.07	5.34±0.35	4.73±0.14	24.91±0.82
Stage 1 SBP 140 -159; DBP 90 – 99	12	1.33±0.23	5.45±0.52	5.28±0.17	28.98±1.46
Stage 2 SBP > 160; DBP > 100	6	1.77±0.65	6.58±0.62	4.52±0.42	27.27±2.13
P – Value		0.525(NS)	0.404(NS)	0.051(NS)	0.018(NS)

n = Number of subjects
S.E.M. = Standard Error of Mean
SBP = Systolic Blood Pressure
DBP = Diastolic Blood Pressure
FPG = Fasting Plasma Glucose
HbA1c = Glycated Haemoglobin
BMI = Body Mass Index
P – Value = Shows level of significance in differences between groups
NS = Not significant (P > 0.05)

Table 7: Pearson Correlation showing relationship between hypertension and Adiponectin.

Correlations			
		Hypertension	**Adiponectin**
Hypertension	Pearson Correlation	1	-.192**
	Sig. (2-tailed)		.010
	N	180	180
Adiponectin	Pearson Correlation	-.192**	1
	Sig. (2-tailed)	.010	
	N	180	180
**. Correlation is significant at the 0.01 level (2-tailed).			

Table 8: Pearson Correlation showing relationship between hypertension and BMI.

Correlations			
		Hypertension	**Body mass index**
Hypertension	Pearson Correlation	1	.297**
	Sig. (2-tailed)		.000
	N	180	180
Body mass index	Pearson Correlation	.297**	1
	Sig. (2-tailed)	.000	
	N	180	180
**. Correlation is significant at the 0.01 level (2-tailed).			

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