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Serum resistin level in diabetes mellitus and at high risk of developing coronary heart diseases in Saudi patients

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Abstract

Background: Recently, human resistin levels have emerged as additional molecular links between obesity, diabetes mellitus and atherosclerosis. The pro- inflammatory resistin contributes to the early stages of atherosclerosis and diabetic complications by promoting endothelial dysfunction and possibly associated with macrophage activation

Aims: The aim of present study is to examine the association between serum resistin level in diabetic patients and at high risk to develop complication in a cross-sectional study of a defined population, taking into account a comprehensive list of risk factors, including Age, Obesity, hyperlipidemia and hypertension.

Study Design: The study is a cross sectional, health facilities based study.

Place, setting and Duration of the Study: The study was conducted at AL Riyadh City, Kingdom of Saudi Arabia (KSA), Diabetic patients whose diagnosed as the WHO classification, during April 2012- September 2013.

Subjects and Methodology: 319 subjects were recruited, 151 of the participants were males and 168 were females. Blood samples were analyzed for biochemical parameters. Serum resistin level was measured for the participants including those who are diabetics and at high risk to develop atherosclerotic cardiovascular diseases.

Results: Serum resistin levels in non diabetic control group were 13.06 ± 9.03 µg/ml lower than that in the group of diabetic patients 18.21 ± 15.42 µg/ml; $P > 0.024$. The level of resistin among diabetic patients with different complication was 20.09 ± 16.97 µg/ml in cardiopathy (n=67); neuropathy (n=25) was 17.73 ± 13.51 µg/ml; nephropathy (n=28) 22.52±20.01 µg/ml; retinopathy (n=38) 20.18 ± 14.83 µg/ml;

hyperlipidemia (n=96) 16.65 ± 15.64 µg/ml and hypertension (n=106) 20.35 ± 17.9 µg/ml. Patient with body mass index ≥ 29 serum resistin level correlated with age, creatnine kinase and CKMB where no correlation with vitamin 25 (OH) D was found in this study.

Conclusion: In this study the level of serum resistin slightly increased in diabetic patients compared to the control group. Nephropathy had higher, whereas hyperlipidemia showed lower resistin level than the other complications. The high level of blood urea nitrogen and hypercreatinemia in renal dysfunction showed correlation with the resistin may indicate its role as a pro-inflammatory that affects the kidney dysfunction. Thought, not deemed significant the increase in serum resistin level in diabetic might indicate a proinflamation in diabetic patients prone to develop cardiovascular diseases.

Keywords: Resistin, Diabetes Mellitus, Renal dysfunction, Coronary heart disease

Introduction

Resistin is an inflammatory cytokines which contributes to the development of atherosclerosis and thereby linked to clinical vascular events. Resistin, is expressed in human macrophages [1], and elevated in subjects with obesity and inflammation [2, 3]. It has direct action on the arterial wall [4, 5] and might be involved in the development of atherosclerosis. Furthermore, several clinical and epidemiological studies have revealed positive associations between plasma concentrations of resistin and proinflammatory cytokines [6-8], which are emerging risk factors for cardiovascular disease. Recently, human resistin levels have emerged as additional molecular links between obesity, diabetes mellitus and atherosclerosis. The pro- inflammatory resistin contributes to the early stages of atherosclerosis and diabetic complications by promoting endothelial dysfunction and possibly associated with macrophage activation. These findings suggest that resistin contributes to the development of atherosclerosis and thereby is linked to clinical vascular events. However, the relationship between resistin and coronary artery disease is highly controversial [9-12]. Furthermore, the information regarding resistin and ischemic stroke in general population is limited to epidemiological study that reported no association between circulating resistin and ischemic stroke [12]. These findings suggest that resistin could explain obesity- related insulin resistance and type 2 diabetes mellitus in human beings. Moreover, resistin can independently predict an individual's risk of heart failure [13]. However, there have been some conflicting reports that do not support a relationship between the blood resistin levels and susceptibility to cardiovascular diseases [8–11]. In addition, not only genetic factors, but also systemic inflammation was suggested to affect the blood levels of resistin [12]. To clarify this controversy, there is clearly a need for more data from different ethnic groups, as serum resistin levels might reflect the impact of genetic or environmental factors on resistin expression and, thus, be subject to ethnic variations [14]. In Saudi Arabia according to national epidemiological study by Al-Nozaha et al., 2004, type-2 diabetes mellitus is increasing at epidemic pace with percent exceeding 25% of the population and its known risk factor for coronary heart diseases. A cross sectional study among patients attending a primary health care clinic revealed 30 % prevalence of diabetes [15, 16] and excess obesity is regarded as the most important risk factor for type 2 diabetes mellitus [17]. Serum resistin level is associated with C-reactive protein and cholesterol in type-2 diabetes in Saudi population [18]. We aim in the present study to examine the association between serum resistin level in diabetic patients with or without complications in a cross-sectional study of a defined population, taking into account a comprehensive list of risk factors, including Age, Obesity, hyperlipidemia and hypertension. Such assessment will predict the individual at high risk to develop complications in the study group.

Subjects and Methodology

This study is a cross sectional, health facilities based study, conducted in central state, AL Riyadh Province, Kingdom of Saudi Arabia, Diabetic patients was diagnosed as Type –1 or 2 Diabetes Mellitus according to the WHO classification

(1999) and fulfilling the set criteria for enrollment. Two hundred and sixty nine diabetic patients (n = 269, 132 males and 137 females) and fifty healthy control subject (19 males and 31 females) were enrolled in the study. 5 ml of blood was collected from the participants in plain vacutainers and serum was separated after centrifugation and stored at -20 C° till analysis.

Demographic and clinical data of the study diabetic groups were collected by a questionnaire from patients and controls prior to samples collection. Weight and height of participants were measured and BMI was calculated. Blood pressure of participants was measured at least three times at 5 min interval. All the necessary precautions to avoid false results were taken.

Laboratory Investigations

All the biochemical measurements were carried out at Faculty of Medicine-Research Laboratory and the Central laboratory, King Fahad Medical City. Blood glucose level was measured from venous blood samples collected following overnight fast using the glucose-oxidase method. Triglycerides and high-density lipoprotein cholesterol (HDL-C) were determined by enzymatic methods (Parsazmun, Karaj, Iran). Serum resistin on fasting blood was measured using a human resistin Quantikine ELISA kit Cat DRSNOO (R & D Systems, UK) following the manufacturer's protocol.

Statistical Analysis

Parametric (mean$\pm$ standard deviation, or standard error of the mean) and nonparametric measurement were calculated using SPSS package version 18 windows. Correlation coefficient and T- test was used and $P \leq 0.05$ was considered as significant.

Results

Out of the 319 subjects included in this study, 199 (62.4%) were type-1 diabetics, 70 (22%) were type-2 diabetics while 50 (15.7) were non diabetic healthy controls. The mean age of the study subject was 40.89 ± 20.26 and the mean duration the diabetes mellitus was 9.37 $\pm$ 7.5 and the body mass index (BMI) was 28.35 $\pm$ 8.44. Serum resistin level in diabetic group was higher (18.21 ± 15.42 µg/ml) than that of control group (13.06 ± 9.03 µg/ml); P value = 0.024. Table 1 shows the characteristics of the subjects. There were significant differences in age, duration of diabetes and BMI. Serum resistin level in patients with type -2 diabetes mellitus was 18.1 ± 15.88 µg/ml whereas in type – 1 diabetes mellitus it was 19.1 ± 12.44 µg/ml, there is difference in the resistin level in type -1 diabetics group compared to the control; P = 0.073.

There was no significant difference in resistin level in relation to BMI in all groups studied, except for diabetics type I with BMI >29, where resistin level was higher than that of the controls.

Table 2 shows the biochemical parameters of lipid profile in the study group. High Density Lipoprotein – Cholesterol (HDL- C) was slightly less in type -2 diabetes mellitus than the type 1 and controls 1.03 ± 0.31, whereas total cholesterol (T. Ch.) in diabetics group was significantly higher than control P = 0.024, also the level of triacylglycerol (TG) in type -2 diabetes mellitus (1.78+ 1.04) was higher than control group (1.14 + 0.61).

Table 3 shows resistin level in co-morbidity among subjects. The highest level of resistin was for nephropathy and the lowest was for neuropathy. There is significant difference in the resistin level between cardiopathy, nephropathy and retinopathy compared to the healthy control.

Table 4 shows the correlation of plasma resistin with biochemical parameters. Resistin level showed significance correlation with the age and duration of the diseases. There is a significant positive correlation with total cholesterol, creatinine and urea. All other parameters including lipid profile were not significantly correlated with plasma resistin.

Discussion

The growing public health concerns surrounding diabetes, obesity and their consequences have focused attention on efforts at treatment and prevention. However, an understanding of the mechanisms underlying these epidemics is essential. Basic and clinical researches have described the inflammatory nature of many metabolic derangements from diabetes to cardiovascular

TABLE 1. Serum resistin µg/ml level and demographic data in the Study group

	Age ± SD	Duration of the disease ± SD	BMI ± SD	Resistin ± SD
Type 2 Diabetes mellitus (n= 201)	53.13 ± 10.76	10.68 ± 8.3	30.76 ± 7.9	18.1 ± 15.8
Type -1 Diabetes mellitus (n= 70)	13.81 ± 6.73	6.73 ± 5.7	21.33 ± 5.6	19.01 ± 12.4
Control (n= 49)	28.41± 9.22		28.32 ± 7.8	13.06 ±9.0
P - value	<0.000 1	0.001	<0.001	0.073

Table 2. Lipid profile and HBA1c **in the study groups**

	HBA1c % ± SD	TG ± SD	T.	LDL - C ± SD	HDL -C± SD
Type 2 Diabetes mellitus	9.15 ± 2.17	1.78 ± 1.04	4.13 ± 1.01	2.49 ± 0.82	1.03 ± 0.31
Type-1 Diabetes mellitus	9.89± 1.89	1.37 ± 1.43	4.45 ± 1.22	2.84 ± .89	1.17 ± 0.44
Control	5.7 ± 0.51	1.14 ± 0.61	3.7 ± 0.17	2.14 ± 0.38	1.19 ± 0.19

Table 3. Resistin level µg/ml in diabetic complications

Cardiopathy N= 67	Neuropathy N= 25	Nephropathy N= 28
20.09 ± 16.97	17.73 ± 13.51	22.52 ± 20.01
Retinopathy N = 38	Hyperlipdemia N = 96	Hypertension N = 106
20.18 ± 14.83	16.65 ± 15.64	20.35 ± 17.9

Table 4. Correlation of resitin level µg/ml with measures of variable biochemical parameters in the study group

Variable	P - value	Variable	P - value
Age	0.038	CK-MB	0.098
DM duration	0.059	CK	0.171
BMI	0.85	LDH	0.148
HBA1c	0.519	Urea	0.011
T. Ch.	.0209	Creatinine	0.005
(LDL-C)	0.104	T3	0.795
(HDL-C)	0.796	T4	0.025
TG	0.448	Calcium	0.319

disease [19]. The hormone resistin, initially described as a rodent adipokine and now understood to be predominantly macrophage-derived in humans is a potential link between inflammation and metabolic disease. Clinical, genetic, and epidemiological studies have strengthened the association between resistin and the prevalence, severity, and outcome of metabolic disease [20, 21]. In this study the level of serum resistin showed slightly increase in diabetic patients compared to the control group. Given the obesity-insulin resistance-inflammation link and convergence of adipocyte and macrophage function, resistin may

provide unique insight into links between obesity, inflammation, and metabolic syndrome risk in humans. However, available data regarding the relationship of serum resistin with BMI and insulin resistance are conflicting. Specifically, AI-Sari et al. confirmed a positive correlation of serum resistin levels with BMI [22]. Tokuyama et al. [23] and Fujinami et al. [24] reported that serum resistin levels in diabetic patients were significantly higher than control, and this change was negatively correlated with insulin sensitivity. Thus, although human resistin is not fat-cell derived, it seems to be well connected to obesity and responsive to changes in adiposity. This connection between macrophage derived resistin and adipose may further the understanding of obesity as an inflammatory state and implicates resistin as a potential modulator of that state. Serum resistin level in the this study showed slightly higher in diabetic complications and and it was higher in nephropathy compared to the other complications, elevated resistin may predict worse outcomes after stroke [25] and appears to correlate with both renal function and CAD risk in those with known end-stage renal disease [26] whereas hyperlipidemia showed lower resistin level compared to other complications, this may reflect the effect of hypolipidemic drugs on circulating resistin in diabetics subject. Whereas resistin level showed no correlation with vitamin 25 (OH) D. Blood urea and creatinine showed significant correlation with the resistin level. The role of resistin as pro-inflammatory adipokines may affect the kidneys dysfunction. Thus circulating resistin levels were associated with general inflammation, renal diseases.

Several groups have investigated the impact of resistin on the development of coronary artery disease (CAD) in humans and not detected a correlation between resistin and either coronary [27, 28, 29]. The disease prevalence or outcome could be due to ethnic variation or to differences in study design and patient selection. Resistin has been most predictive in prospective and cohort studies which do not exclude those with known or prior CAD and may have less confounding due to medical therapy or other biases. In this study the there is a positive correlation between resistin and cardiac marker CK-MB. It has become clear that there is a potential connection between resistin and CAD outcomes across a spectrum of populations. The role of resistin as an independent prognostic biomarker remains unclear; the impact of resistin on CAD and other heart disease is gaining attention.

Ethical Consideration

The project was reviewed and approved by the Institutional Review board – IRP King Fahad Medical City, Log number 11-040. All patients were consented before their enrollment in the study.

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