

Serum Adiponectin and Vaspin levels in Abdominal Obesity and Type 2 Diabetes Mellitus

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Abstract

Objective: Adiponectin and vaspin are adipocyte-derived proteins and involved in the regulation of glucose metabolism and obesity related disorders. The mechanisms that adipokines might be linked to disturbance of glucose are unclear. Thus, this study investigated the levels of serum adiponectin and vaspin, blood lipid profiles, and abdominal obesity in type 2 diabetes (T2DM) patients and controls.

Materials and Methods: Frothy T2DM patients and 40 healthy volunteers matched by age and body mass index (BMI) were enrolled in the study. The levels of serum adiponectin, vaspin, fasting blood glucose (FBG), 2-hr post prandial (2hr-PP), and lipid profile were measured in both groups. Anthropometric parameters including BMI, waist circumference (WC) and waist to height ratio (WHR) were measured.

Results: Higher levels of vaspin and lower levels of circulating adiponectin were observed in T2DM patients than in the controls (P -value:0.0001). In multivariate regression analyses adjusted for age and BMI, serum adiponectin levels showed a significant negative correlation with WC, WHR, FBG and duration of diabetes, and a positive correlation with HDL. While, the vaspin levels were positively correlated with WC, WHR and duration of diabetes in T2DM patients. In addition, vaspin and adiponectin levels had negative correlation with each other.

Conclusion: The findings suggested that the abdominal obesity had the highest relationship with adiponectin and vaspin in diabetic patients.

Keywords: Adiponectin, Vaspin, Obesity, Type 2 diabetes

Introduction

The prevalence of type 2 diabetes mellitus (T2DM) is increasing worldwide (1). Diabetes mellitus is one of the pathophysiological features of the metabolic syndrome, which may leads to high risk of cardiovascular disease (2). Several

lifestyle-related risk factors were documented in developing T2DM including, atherosclerosis, hypertension, dyslipidemia, abdominal obesity, family history of T2DM, ageing, history of gestational diabetes mellitus, insulin resistance, and sedentary lifestyle (1,3).

In addition to the classic risk factors, recent studies demonstrated that some adipocyte-derived hormones such as adiponectin and vaspin (visceral fat tissue- derived serpin) are also involved in obesity related disorders (1,4-6).

Adiponectin, a novel protein derived from adipose tissue, is a major protective factor which acts against metabolic and cardiovascular consequences of obesity (7). Adiponectin has anti-inflammatory, anti-atherogenic and insulin-sensitizing effects (1,3). However, there are controversial findings about the association of adiponectin and T2DM. (1,3,8-10)

Vaspin is a small protein (~50 kDa) of the adipokines family which was firstly isolated from visceral white adipose tissues (WATs) of Otsuka Long-Evans Tokushima Fatty (OLETF) rats with T2DM (11-12). Vaspin is also secreted from the central nervous system (especially the hypothalamus), skin, pancreatic islets and stomach (13). Numerous studies demonstrated that the vaspin is associated with metabolic risk factors such as obesity, insulin resistance and T2DM (11,13,14). The variation in levels of vaspin was demonstrated in different studies. In a study, no difference in the levels of serum vaspin between individuals with normal glucose tolerance and T2DM was found (11). Another study reported that the serum vaspin levels were lower in T2DM patients compared with non-diabetic subjects (8). In some studies, circulating vaspin in the obese patients with T2DM was higher than normal body mass index (BMI) controls (14-16). Due to high prevalence of obesity in T2DM patients and conflicting data regarding the relationship between adiponectin, vaspin and T2DM, the aim of this study was to compare serum levels of adiponectin and vaspin, blood lipid profiles and obesity in patients with T2DM and controls.

Materials and Methods

This analytical cross-sectional study was performed on 40 T2DM patients (between 31-78 years old and BMI range of 22.7- 49

kg/m²), referred to the diabetes clinic at Ali-Asghar hospital affiliated to Zahedan University of Medical Sciences (Zahedan, Iran). Forty non-diabetic volunteers, matched for age and BMI were selected as control group (aged 30-75 years and BMI range of 21.3-44 kg/m²). The study was performed during August to December 2015. Non-diabetic subjects were defined as fasting blood glucose (FBG) <126 mg/dL and a 2-hr post prandial (2hpp) <140 mg/dL (17). Exclusion criteria were; inflammatory disease, clinical signs of infection, medical history of anti-hypertensive or blood lipid lowering drugs, pregnancy, cardiovascular disease, thyroid disorders, smoking or alcohol abuse.

Blood samples were taken from all subjects after a 14hours overnight fasting and immediately frozen in aliquots at -70°C until analysis. For evaluation of FBG and 2hpp, blood samples were measured at baseline and 2 hours after high carbohydrate diet using an automatic analyzer (Hitachi, Japan). Serum levels of total cholesterol (TC), triglyceride (TG), high- density lipoprotein cholesterol (HDL-C), low- density lipoprotein cholesterol (LDL-C) were measured by the commercial kits (Pars Azmoon ,Tehran, Iran) using the automatic analyzer (Hitachi, Japan).

Serum levels of vaspin and adiponectin were measured using Human ELISA kits (Visceral adipose-specific serine protease inhibitor); Hangzhou East biopharm CO., LTD. Cat .No: CK-E10968), and Human Adiponectin ELISA kit (Boster Biological Technology .Cat No: EK0595, USA), respectively.

Weight and height participants were measured by Seca scale with normal clothing and without shoes. BMI was calculated as weight in kilograms (kg) divided by the square of the height in meters (m²), and was categorized according the World Health Organization recommendations; BMI>25 Kg/m² were considered as overweight/obese (17).

The waist circumference (WC) and waist to height ratio (WHR) were measured as simple screening tools for measuring abdominal obesity. Waist circumference was measured midway between the lower rib margin and the

iliac crest at the end of a gentle expiration. WC>102 in men, WC>88 in women and WHR > 0.5 for both men and women were considered as abdominal obesity (17,18).

This study was approved by the ethics committee of Zahedan University of Medical Sciences (approval date 4 July 2015; number 7330). All subjects gave informed consent before taking part in the study.

Statistical analysis

Data were analyzed using SPSS statistical software package program (version 18 for windows, Chicago, USA). The results were expressed as mean \pm standard deviation (SD) with range, as appropriate. The data were checked for normal distribution using the one-sample Kolmogorov-Smirnov test, and were compared by the student's T-test. Pearson

correlation coefficient was used for assessment of correlations. Multivariable regression analysis was performed to detect the risk factors of T2DM development, with adjustment for potential confounding factors—value < 0.05 was considered significant.

Results

Demographic and biochemical characteristics of patients with T2DM and controls were summarized in Table 1. There were no significant differences between two groups in BMI, age, gender and HDL-C levels.

Adiponectin

Adiponectin serum concentrations ranged from 1-25.4 (mg/L). In T2DM patients circulating adiponectin was significantly lower in comparison with control subjects (10 ± 3.3

Table 1. Characteristics of studied subjects

Groups Parameters	T2DM Patients	Controls	P-value
Age (yrs)	52.7 \pm 10.6	50.2 \pm 10.6	0.28
Weight (kg)	70 \pm 12.7	72 \pm 10.3	0.43
BMI (Kg/m ²)	28.4 \pm 4.7	27 \pm 3.2	0.137
WC (Cm)	134.3 \pm 13	83.7 \pm 14.3	0.0001
WHR	0.68 \pm 0.1	0.52 \pm 0.08	0.0001
FBG (mg/dL)	214 \pm 86	84 \pm 7.7	0.0001
2 hr-PP (mg/dL)	290.5 \pm 112	93.2 \pm 10.5	0.0001
Cholesterol (mg/dL)	187 \pm 51	159 \pm 32	0.004
TG (mg/dL)	210 \pm 102	85 \pm 56	0.0001
LDL-C (mg/dL)	109 \pm 43	81 \pm 18	0.0001
HDL-C (mg/dL)	48 \pm 9.3	47.5 \pm 20.5	0.89
Vaspin to Adiponectin Ratio	0.29 \pm 0.06	0.092 \pm 0.01	0.001

Data were shown by mean \pm SD.

BMI: body mass index; WC: waist circumference; WHtR ;waist to height ratio; FBG: fasting blood glucose ; 2 hr- PP: 2 hour –post prandial ; LDL-C: low density lipoprotein -cholesterol; HDL-C: high density lipoprotein-cholesterol.

Table 2. Multivariate linear regression analysis between adiponectin and vaspin and different parameters in T2DM patients

Parameters	Standardized Coefficients			
	Adiponectin		Vaspin	
	β -Coefficient	P-value	β -Coefficient	P-value
BMI	-0.113	0.538	0.252	0.280
WC	-0.330	0.044	1.129	0.035
WHR	-0.456	0.010	0.730	0.049
Duration	-0.535	0.000	0.889	0.000
FBG	-0.259	0.009	0.169	0.068
2hr-PP	-0.153	0.587	0.301	0.426
Cholesterol	-0.202	0.725	0.703	0.809
TG	-0.291	0.277	0.209	0.963
LDL	-0.005	0.993	0.512	0.302
HDL	0.338	0.038	0.869	0.392

BMI: body mass index; WC: waist circumference; WHtR: waist to height ratio; FBG: fasting blood glucose; 2 hr-PP: 2 hour – post prandial; TG: triglyceride; LDL-C: low density lipoprotein -cholesterol; HDL-C: high density lipoprotein- cholesterol.

(mg/L) vs. 15.3 ± 6.6 (mg/L), respectively (P -value: 0.0001) (Fig 1).

The Pearson correlation revealed significant negative correlations between serum adiponectin levels and WC ($r = -0.42$, P -value: 0.008), WHR ($r = -0.48$, P -value: 0.002), FBG ($r = -0.40$, P -value: 0.01), 2 hpp ($r = -0.47$, P -value: 0.003), TG ($r = -0.94$, P -value: 0.01) and duration of diabetes ($r = -0.535$, P -value: 0.0001), and a significant positive correlation with HDL-C ($r = 0.98$, P -value: 0.004) in T2DM patients.

In addition, in multivariate regression analysis adjusted for age and BMI, serum adiponectin

levels showed a significant negative correlation with WC ($\beta = -0.330$, P -value: 0.05), WHR ($\beta = -0.456$, P -value: 0.01), FBS ($\beta = -0.259$, P -value: 0.01) and duration of diabetes ($\beta = -0.535$, P -value: 0.0001), and a positive correlation with HDL-C ($\beta = 0.338$, P -value: 0.05) in T2DM patients (Table 2).

Vaspin

Vaspin serum concentrations ranged from 0.6-7.3 ng/mL. Serum vaspin levels were markedly higher in the T2DM patients compared with healthy controls (2.1 ± 1.5 (ng/mL) vs. 1.4 ± 0.6 (ng/mL), respectively

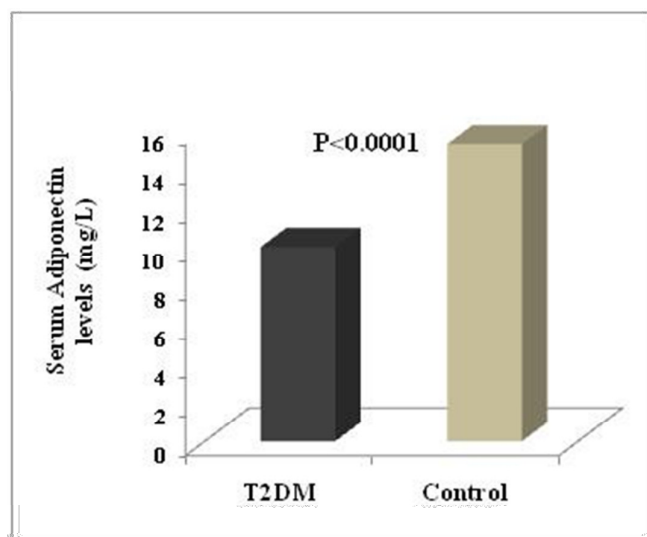


Figure 1. Adiponectin serum levels in T2DM patients and control subjects

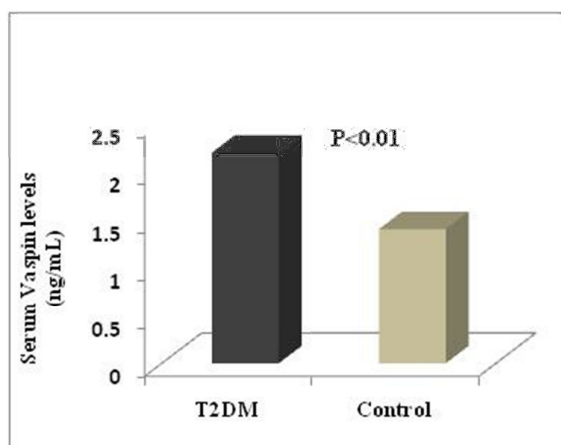


Figure 2. Vaspin serum levels in T2DM patients and control subjects

(*P*-value: 0.01)) (Fig 2).

The Pearson correlation revealed a significant positive correlations between serum levels of vaspin and WC ($r= 0.48$, *P*-value: 0.002), WHR ($r= 0.36$, *P*-value: 0.02), FBS ($r= 0.43$, *P*-value: 0.051) and duration of diabetes ($r= 0.89$, *P*-value: 0.0001) in T2DM patients.

In addition, vaspin and adiponectin levels were negatively correlated ($r=-0.476$, *P*-value: 0.01).

In multivariate analyses, significant positive correlations were detected between the vaspin levels and WC ($\beta= 0.929$, *P*-value: 0.05) and WHR ($\beta= 0.730$, *P*-value: 0.05) and duration of diabetes ($\beta= 0.889$, *P*-value: 0.0001) (Table 2).

Discussion

T2DM is a common metabolic disorder in the world with 7.7% prevalence in Iranian adults (2). Obesity is one of the most important risk factors of T2DM that is associated with decreased insulin sensitivity, dyslipidemia and hypertension (19,4). Obesity and abdominal obesity, as the most common risk factors in the prediction of T2DM were reported in two recent decades in Iran (18,19). Our results also demonstrated that T2DM patients had significantly higher WC and WHR in comparison with controls. Thus, the assessment of abdominal obesity and levels of adipocytokines secreted by adipose tissue may be useful for high risk group identification (1,3,6).

The circulating concentrations of adiponectin decrease according with insulin resistance during development of T2DM (1,8,20). Also it is associated with some features of the metabolic syndrome, such as abdominal obesity, dyslipidemia, hypertension, gender, aging and lifestyle (1,20,21).

The results of current study also indicated that T2DM patients had lower adiponectin levels than controls. Adiponectin serum level decreases the worsening of diabetes. Furthermore, a negative association between serum adiponectin and obesity, particularly those with abdominal obesity, blood glucose

and triglyceride, and a positive correlation with HDL-C was found in T2DM patients. This study findings suggested that low adiponectin concentration may be an independent risk factor of T2DM and metabolic syndrome (1,9,10,22-24).

The present study demonstrated that T2DM patients with abdominal obesity and hypertriglyceridemia had lower adiponectin levels than patients who did not (21,24-26). Moreover, decrease of circulating adiponectin in T2DM patients was found to be markedly associated with longer duration of diabetes, according with previous report (21). This suggests that adiponectin may be associated with duration of T2DM.

Although, the association between adiponectin concentrations in human obesity and T2DM is unknown, but adiponectin increases energy utilization and fatty-acid consumption, which leads to a reduction in the liver and skeletal muscle triglyceride contents, and thus directly regulates glucose metabolism and insulin sensitivity (27). On the other hand, some sexual hormones, such as estrogen and testosterone, were also suggested in the regulation of plasma adiponectin level (21,26), but we found no sexual difference in circulating adiponectin, and future studies are required. In overall, reduced adiponectin levels in T2DM patients might be linked to metabolic syndrome and adverse cardiovascular events.

In addition, other adipokines were also found to contribute the progression of diabetes and metabolic disorders (13). Numerous studies evaluated the vaspin levels in T2DM patients and vaspin is associated with obesity, hyperlipidemia and T2DM (8,11,12,14,15,28). In our study, it was found that serum vaspin levels were markedly higher in T2DM patients than control. Significantly higher circulating vaspin levels were observed in T2DM patients with abdominal obesity compared with non-obese T2DM patients and controls, which were accordant with some other studies (11-15). This association persisted after multivariate adjustment.

In diabetic patients, it was also found that increased circulating vaspin level was associated with elevated glycemic levels and long duration of diabetes. However, after adjusting for age, sex and BMI, circulating vaspin did not correlate with FPG, whereas the positive correlation with duration of diabetes persisted in multiple regression analysis. It is possible that over a long period of diabetes and abdominal fat accumulation in T2DM patients, the levels of serum vaspin is increased

Another studies detected that the longer period of diabetes, insulin treatment and insulin resistance in obese patients with T2DM may affect the vaspin levels (4,8,28). In our study all patients were on diabetic oral medication.

Furthermore, a negative association between serum levels of vaspin and adiponectin was in T2DM patients, and this correlation persisted after adjusting for age and BMI.

The reported relationships between serum vaspin and adiponectin were controversial (8). In a study performed in obese children and adolescents, vaspin levels were negatively correlated with adiponectin levels (29). In another study, a significant positive correlation was found between serum vaspin and adiponectin levels in childbearing age women (28).

The results of studies indicate that the relationship between serum adiponectin and vaspin levels can be affected by sex, age, body weight and other metabolic status (21,28,29). However, no age and gender differences in vaspin serum concentrations were found in our study.

Furthermore, in the current study, a significant reduction of vaspin to adiponectin ratio was demonstrated in T2DM patients, and the vaspin to adiponectin ratio was more strongly

correlated with WC and WHR than vaspin or adiponectin levels alone.

The main limitation of the present study is its methodology, as it is a cross-sectional study with small sample size. Moreover, we did not measure the insulin resistance levels in the T2DM patients. Thus, additional studies are necessary to validate our results.

Conclusions

The findings suggested longer duration of diabetes and abdominal obesity had the highest correlation with adiponectin and vaspin in diabetic patients. Due to the high prevalence of obesity in diabetes patients, the assessment of these markers reflects body fat mass, and may be able to predict the high risk groups.

Authors' contributions

Dr. Montazerifar and Dr. Karajibani contributed in designing of study, drafting of manuscript, analysis, and discussion. Dr. Kheykhaie contributed in diagnosis of diabetes. Mohammadi and Hemmat Jouy contributed in data collection and Rezaie contributed in testing.

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