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RELATIONSHIP OF ADIPONECTIN WITH INSULIN RESISTANCE, LIPID PROFILE AND BODY MASS INDEX

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Abstract

Adiponectin is a fat derived protein. It is linked to obesity and its co-morbidities especially insulin resistance, type 2 diabetes, and certain components of metabolic syndrome.

Objective: The objective of this study was to correlate serum adiponectin levels with Insulin sensitivity, Lipid Profile and with BMI in self-reported healthy individuals. and to evaluate the relationship between serum adiponectin levels and key metabolic parameters including insulin resistance (HOMA-IR), lipid profile, and body mass index (BMI) in self-reported healthy individuals from the Pakistani population.

Methods: We designed a cross-sectional analytical study and recruited a cohort of eighty (80) self-reported healthy individuals in the study by non-probability convenient sampling. The whole experimental work was conducted at the Institute of Molecular Biology and Biotechnology (IMBB), The University of Lahore from September 2021 to June 2022. The adiponectin levels in patients' serum were assessed by ELISA kit. Lipid profile was assessed by commercial Human Diagnostics® kits.

Results: Our results showed that mean value of serum adiponectin levels in subjects was 5.55±1.93 (Mean±SD) with no significant difference between male (5.43± 1.35) and female subjects (5.75 ± 2.59) at p>0.50. There was no significant correlation between adiponectin and any of the anthropometric or metabolic parameters including waist circumference, BMI, blood sugar fasting, serum insulin, Homeostatic Model Assessment for Insulin Resistance (HOMA-IR) and lipid profile at p>0.50 when checked by Spearman correlation testing. However, when multiple linear regression was used to check the association of adiponectin levels with waist circumference while controlling age and serum lipids, a statistically significant link was revealed (beta=0.34, p=0.03).

Conclusions: Our study adds to the developing pool of knowledge about the physiological role of adiponectin. There is no simple correlation of this adipokine with anthropometric and metabolic parameters. A positive association with waist circumference may be linked to the visceral intraabdominal fat which is a source of this adipokine. More studies are required to find out physiological role of adipokines on genetic and molecular grounds.

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Introduction:

The adipose tissue is a fascinating organ that contains within itself an array of secretions that regulate and affect different processes that are vital for the survival and adequate functioning of the human body. The white adipose tissue is responsible for releasing adipokines, the biologically active compounds behind the many actions of the adipose tissue¹. Adiponectin is a 244-amino-acid protein that resembles collagen. It is present in the blood at levels ranging from 5-30µg/mL (5000-30,000 ng/mL). This versatile polypeptide hormone has wide systemic effects with its levels inversely related to insulin resistance and body fat². Adiponectin works via AdipoR1 and AdipoR2 receptors. AdipoR1 is mostly located in skeletal muscle, while AdipoR2 is substantially found in the liver, carrying out processes affecting carbohydrate and lipid metabolism¹.

The term "Metabolic Syndrome" does not refer to a single disease entity but a combination of various predisposing features and distinctive clinical characteristics that increases the possibility of developing cardiovascular problems, diabetes, cerebrovascular hemorrhage and a multitude of various other medical issues. The diagnosis is confirmed when at least three of the five biological threats are imposed on the body. These threats include elevated levels of blood glucose and triglycerides with reduced levels of HDL ("good") cholesterol in the circulation, hypertension and an outsized waist circumference, usually described as an "apple-shaped" body. Of the five threats, central obesity is the most significant factor resulting in metabolic syndrome³.

Insulin resistance is a complex phenomenon but can be simply described as a decreased response of body tissues and cells to insulin, which leads to a deranged control of glucose and lipids in the blood. This is especially expressed in a Diabetes Mellitus Type 2 (T2DM), a disease principally defined by the faulty glucose control in the body secondary to a decreased biological response to insulin. According to research, adiponectin may have a pathogenic role in T2DM. Obese diabetics were found to have significantly reduced adiponectin levels. It has been hypothesized that by way of AdipoR1 and AdipoR2 receptors, low levels of adiponectin caused a reduced uptake of glucose in the skeletal muscle, reduced the capability of insulin to suppress lipolysis in adipose cells, diminished insulinmediated inhibition of gluconeogenesis in the liver, and hindered secretion of insulin in the pancreas. All causing impaired glucose metabolism and increased insulin resistance⁴. This is also reflected by increased HOMA-IR levels (Homeostatic Model Assessment of Insulin Resistance) in diabetic obese individuals⁵.

Decrease levels of adiponectin are associated with dyslipidemia (increased LDL, VLDL, TG and decreased HDL levels). Insulin resistance caused by decreased levels of adiponectin is associated with excessive free fatty acids, which are transformed to VLDL (very low-density lipoprotein) in the liver. This hypertriglyceridemia causes atherogenesis due to accelerated LDL (low-density lipoprotein) production along with excessive HDL (high-density lipoprotein) removal^{6,7}.

Abdominal fat is known to be closely linked to metabolic syndrome and heart diseases. Fat, in the form of fatty acid, is shunted into the portal circulation and this directly affects the waist diameter. Adiponectin levels have been shown to be low in obese people, which may be the cause of insulin sensitivity and add to metabolic syndrome⁸.

Adiponectin is a product of fat cells in the body but its levels are paradoxically suppressed in states of obesity, a trait shared by none of the adipokines. This exposes the body to an inflammatory condition without the protective actions of adiponectin^{9, 10}. Among many tools developed to classify obesity, Body Mass Index is a simple and an efficient method of labeling the amount of obesity and to simplify the risk an individual may have in developing certain illnesses, with waist-to-hip ratio being the chief indicator¹¹. Increased BMI is associated with decreased levels of adiponectin^{12,13} with adiponectin having beneficial effects while its decreased levels being associated with various metabolic abnormalities¹⁴.

The data related to the adiponectin is limited in Pakistani population. The current study aims to investigate the relationship of adiponectin with various indicators of metabolic syndrome in healthy individuals and thus sheds light on the physiology of the adiponectin.

methods:

This cross-sectional comparative study was conducted at the institute of Molecular Biology and Biotechnology, The University of Lahore, from September 2021 to June 2022 after obtaining approval from Institutional Review Board of The University of Lahore (Ref-IMBB/BBBC/21/627). Mean concentrations of adiponectin in obese patients were 14.6 μg/ml and 18.9 μg/ml in non-obese controls¹⁵. Mean standard deviation (S.D) was 2.15 and variance was 4.6. Confidence interval was taken as 95. Power was taken as 80. The estimated sample size was 4 but for the better power of study we took a sample of 80.

Total of 80 self-reported healthy volunteer from general population fulfilling the inclusion criteria took part in this study. Pregnant females, individuals with less than 18kg/m² BMI and patients with chronic illness were excluded from this study. Informed consent was obtained by all screened patients who filled out the questionnaire for their demographic details, height and weight for BMI calculation, waist-circumference as well as risk factor history. 10.0 ml blood sample was taken from the accessible vein under aseptic measures.

Blood was further processed for the estimation of fasting glucose levels, insulin levels, lipid profile and adiponectin in the samples collected from selected patients. Samples (blood) were taken in EDTA tubes. After centrifugation of blood at 4000 rpm for 10 minutes, serum was separated. The levels of cholesterol, triglycerides, low-density lipoproteins (LDL), high-density lipoproteins (HDL), very-low density lipoproteins (VLDL) were estimated in the samples of serum by using commercial Human Diagnostics® kits. Blood Insulin concentration levels were estimated by using Abbott kit in the lab. The adiponectin was assessed by Adiponectin ELABSCIENCE ELISA® kit. Insulin resistance was calculated by using the HOMA-IR equation: HOMA-IR = Insulin (μ U/ml) X Glucose (mg/dl)/ 405^{16} .Data was entered in SPSS version 21.

The correlation between adiponectin and any of the anthropometric or metabolic parameters including waist circumference, body BMI, blood sugar fasting, serum insulin, Homeostatic Model Assessment for Insulin Resistance (HOMA-IR) and lipid profile at p>0.50 was checked by Spearman correlation testing. Multiple linear regression was also used to check the association of adiponectin levels with waist circumference while controlling age and serum lipids. Results were presented as mean \pm SD. p value ≤ 0.005 was considered significant and p value ≤ 0.001 was considered highly significant.

RESULTS:

The demographic data of the subjects is shown in table 1. There were 48 males (54%) and 32 females (46%) with mean ages of 34.47 ± 12.29 and 33.00 ± 21 years respectively. Table 1 is showing the characteristic

Table 1: Demographic and baseline characteristics of the participants (n=80)

Sr.#	CHARACTERISTIC	FREQUENCY	PERCENTAGE
1	Gender	Male: 48	54 %
		Female :32	46 %
2	Age in years*	34.47 ± 12.29	33.00 ± 21.00
3	Body Mass Index in kg/m2*	27.34 ± 7.47	26.90 ± 15.00
4	Waist Circumference inches*	38.61 ± 12.055	36.00 ± 19.00
5	Fasting blood sugar in mg/dl	105.05 ± 34.95	95.19 ± 39.20
6	Serum insulin in uIU/ml	44.62 ± 65.56	21.80 ± 44.25
7	HOMA-IR	34.17 ± 56.64	13.30 ± 34.24
8	Serum adiponectin in ng/mL	5.59 ± 1.94	5.34 ± 2.54

Table 2: Serum Lipid profile levels in male and female subjects

SR#	LIPID PROFILE	Males	Females	'P'*
1	CHOLESTEROL in mg/dl ^a	167.41±39.94	179.60±46.29	0.49
2	HDL in mg/dl	35.58±13.82	42.77±10.63	0.01 ^b
3	LDL in mg/dl	86.62±26.01	96.45±50.08	0.11
4	VLDL in mg/dl	30.69±26.01	33.10±38.48	0.82
5	TRIGLYCERIDES in mg/dl	164.69±130.84	175.33±182.57	0.82

^{*}p value for independent samples't' test or Mann Whitney test to test difference in means or medians as applicable

Mean levels of serum lipids including LDL, VLDL, TGs and free cholesterol in males and females are shown in tables-2. As can be seen in the table-2, serum cholesterol is normally distributed (Shapiro Wilk test p=0.64 while LDL, VLDL, HDL and TGS are skewed (p<0.05). There are no significant differences in serum lipid levels between males and female subjects (p>0.05) except HDL, in which case HDL levels are higher in females as compared to males (p=0.01). Table 2 contains the statistical values

Mean adiponectin levels in the subjects were 5.59 ± 1.94 ug/mL. The levels in male subjects were 5.42 ± 1.35 ug/mL) whereas in females these were 5.74 ± 2.58 but this difference was not statistically significant (p=0.47). As shown in figures 1 and 2, mean serum adiponectin did not differ significantly between waist circumference groups (p>0.05) or age groups (p>0.05)

Spearman correlation between serum adiponectin levels and various anthropometric and metabolic parameters revealed no significant correlations. There was inverse correlation of between adiponectin and age (r= -0.06, p>0.05), serum insulin (r=-0.09, p>0.05) and fasting blood sugar (r= -0.198, p>0.05), but these did not reach statistical significance. The correlation between serum adiponectin and gender (r=+0.001), BMI (r=+0.10), waist circumference (r=+0.17), HOMA-IR (r=0.06), Cholesterol (r=+0.13), HDL (r=+0.13), LDL (r=+0.08), VLDL(r=+0.14) and triglycerides (r=+0.04) were positive but again did not reach statistical significance (p>0.05). This is shown in Table-3. However, when we adjusted for serum insulin and serum LDL by multiple linear regression modeling, the statistically significant association of adiponectin with waist circumference emerges (Beta=+0.34, p=0.04) as shown in table-4.

Table 3: Spearman correlation showing relationship between adiponectin levels and various metabolic and anthropometric variables.

Sr. #	Variable	Correlation Coefficient (r)	P value
1	Age (years)	- 0.06	0.60
2	Gender (M/F)	+ 0.001	0.96
3	Body Mass Index kg/m ²	+ 0.10	0.37
4	Waist Circumference(inches)	+ 0.17	0.16
5	Fasting Blood Sugar (mg/dl)	- 0.198	0.08
6	Serum Insulin Level (fasting) (uIU/ml)	- 0.09	0.42
7	HOMA-IR	+ 0.06	0.60
8	Cholesterol (mg/dl)	+ 0.13	0.25
9	HDL (mg/dl)	+ 0.13	0.26
10	LDL (mg/dl)	+ 0.08	0.47
11	VLDL(mg/dl)	+ 0.14	0.23
12	Triglycerides (mg/dl)	+ 0.04	0.75

^a mean level \pm SD are shown for cholesterol where median levels \pm IQR are shown for other lipids ^b significant difference between medians as shown by Mann Whitney U test

Table 4: Multiple linear regression association of waist circumference with adiponectin after adjusting for serum LDL and serum insulin

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Variables	Unstandardized		Standardized	Sig.	95.0% Confidence	
	Coefficients		Coefficients		Interval for B	
	В	Std. Error	Beta		Lower	Upper
					Bound	Bound
Waist Circumference	0.42	0.20	0.34	0.04*	0.03	0.81
inches						
Serum LDL mg/dL	-0.01	0.14	-0.01	0.9	-0.29	0.27
Serum Insulin mg/dL	-0.07	0.05	-0.21	0.16	-0.16	0.03

^{*} Statistically significant

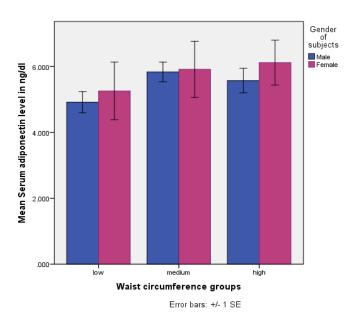


Fig 1: Mean serum adiponectin levels distribution according to waist circumference groups (p>0.05)

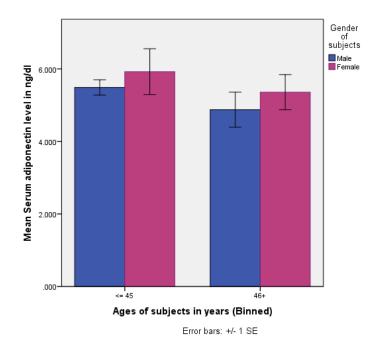


Fig 2: Mean serum adiponectin levels distribution according to age groups (p>0.05)

DISSCUSSION

The current study reports serum adiponectin levels in physiologically healthy subjects from Pakistani population. The average serum adiponectin levels were 5.59± 1.94 ug/mL; 5.42±1.35 ug/mL in men and 5.74±2.58 ug/mL in women. There was a significant positive association of serum adiponectin with waist circumference after adjusting for LDL and age (Beta= +0.42, p=0.04). There was no significant correlation of adiponectin with lipid profile including cholesterol, triglycerides, LDL, VLDL, HDL, BMI and insulin resistance (HOMO-IR and serum insulin levels).

In a study conducted by Aisike et al., 178 subjects from previous cross-sectional surveys were recruited and divided into 3 groups (healthy group, metabolically healthy obesity, metabolically unhealthy obesity) based upon BMI and diagnostic criteria of metabolic syndrome. 'Metabolically unhealthy obesity' group had high BMI, waist-hip ratio, waist circumference, total cholesterol, LDL, triglycerides, fasting plasma glucose levels, Homa-IR levels and blood pressure while serum HDL levels were significantly lower in this this group. Adiponectin had a positive correlation with systolic blood pressure and negative correlation with fasting blood sugar levels in metabolically healthy group. In 'metabolically unhealthy group' adiponectin had a positive correlation with total cholesterol and systolic blood pressure. Linear regression analysis showed that systolic blood pressure, triglycerides, LDL were independently correlated with adiponectin while BMI was independently correlated with leptin in obese subjects 17. In our study, we evaluated the correlation of adiponectin levels with age, gender, BMI, waist circumference, fasting blood sugar, HOMA-IR, insulin levels, lipid profile. We found no significant correlation except for waist circumference and adiponectin levels on multiple linear regression analysis. Our results are different from this study because we had small sample size. Further studies might be planned in which individuals are categorized into groups based on their BMI and those groups are compared for adiponectin levels, lipid profile, fasting blood sugar levels, insulin resistance, and HOMA-IR.

In a study conducted by Munhoz et al., diluted plasma of obese humans when injected to pancreas invitro impaired beta cell function and insulin secretion while plasma of lean individuals had opposite outcome. Similar results were obtained with sera of obese rats when compared to sera of lean rats. Increased levels of adiponectin were found in the serum of lean individuals/ rats, and these were found to have protective effect on beta cell functionality. When adiponectin was administered to the beta cells treated with serum of obese donors, their functionality was completely restored. This study highlighted that decreased levels of adiponectin were responsible for beta cell dysfunction¹⁸. In our study, we found no statistically significant correlation of adiponectin with obesity, lipid levels, insulin resistance and fasting glucose levels. We need to study a large population with cohorts from different age groups and categorize those individuals on the basis of BMI into 3 groups (normal BMI, obese, morbidly obese) to see the correlation of BMI with adiponectin, insulin resistance, fasting glucose levels and lipid levels.

In a study conducted by Becerril et al., effect of age and sex on obesity and development of type-II diabetes was evaluated. Male and female mice were fed normal diet or high fat diet for 12 or 32 weeks. Female mice showed significantly less changes in body weight, body weight gain and adiposity index as compared to male mice. Aged female had high adiponectin/leptin ratio which was negatively correlated with body weight, fat deposits and insulin resistance. This study indicates that females are more protected from obesity and its comorbidities as compared to males with adiponectin being the key player 1 our study, we could not find any correlation of adiponectin with gender. In our study, females were less in number as compared to males (54% males, 46% females). We need a large sample size with 1:1 ratio of males and females.

In a cross-sectional survey conducted by Hong et al., 3680 individuals aged 18-70 years were evaluated for the association of adiponectin levels and the risk for development of type-II diabetes. There was negative association between adiponectin levels and type-II diabetes in a population with less risk factors for development of type-II diabetes, but this relationship gradually decreased with accumulation of more risk factors for development of type-II diabetes like hypertension, dyslipidemia, etc²⁰. Our results were contradictory to this study and we found no significant correlation of adiponectin to fasting sugar levels, insulin resistance, or BMI perhaps because of small sample size.

We recruited healthy individuals only, we need to add individuals with some of the features of metabolic syndrome to our study. The correlation might be different in healthy subjects and subjects with some metabolic abnormalities.

Conclusions

There is no simple correlation of adiponectin with anthropometric and metabolic parameters. A positive association with waist circumference may be linked to the visceral intraabdominal fat which is a source of this adipokine. More studies are required to find out physiological role of adipokines on genetic and molecular grounds.

Author contributions

Conflict of interest: None

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