

Association of serum leptin levels with body mass index, C- reactive protein and homocysteine in patients with type 2 diabetes mellitus

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ABSTRACT

Introduction: Diabetes mellitus (DM), one of the most common global endocrine disorders is emerging as a growing health care problem. The risk of cardiovascular diseases is several folds higher in type 2 diabetics as compared to normal subjects.

Objectives: To determine the association of leptin with BMI, C-reactive protein and homocysteine in type 2 diabetics.

Materials & Methods: It was a cross sectional study conducted in Hayatabad Medical Complex (HMC) and Rehman Medical Institute (RMI), Peshawar. Study duration was six months. A total of 96 type 2 diabetics were enrolled in the study after taking informed consent. Serum leptin levels were determined and plasma levels of hs-CRP and homocysteine were measured. Data were analyzed by SPSS version 20. A p-value of ≤ 0.05 was taken as significant.

Results: Out of 96 subjects, 23 were males and 73 were females. The correlation of serum leptin with BMI was positive and significant ($r=0.418$, $p=0.01$). The correlation with HbA1c was also significant and positive ($r=0.207$, $p=0.04$). The correlation with duration of diabetes was also significant and positive ($r=0.223$, $p=0.03$). The correlation with hs-CRP was positive but not significant ($r=0.249$, $p=0.467$). The correlation with homocysteine was also positive but not significant ($r=0.348$, $p=0.10$).

Conclusion: There is a positive association between serum leptin levels and BMI. The study also concluded that there is a positive association of cardiovascular risk markers with serum leptin levels.

Keywords: Type 2 Diabetes Mellitus; Leptin; C-Reactive Protein; Homocysteine; BMI.

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INTRODUCTION

Diabetes mellitus (DM) is one of the most common endocrine disorders and is a growing health care problem all over the world¹. Type 2 diabetes mellitus occurs in 90-95% of individuals with diabetes and is also called adult onset diabetes or non-insulin dependent diabetes. In these individuals there is insulin resistance and only relative insulin deficiency². It is characterized by hyperglycemia resulting in damage to various organ systems like blood vessels, eyes, nerves, kidneys and even affecting the gastrointestinal tract¹.

The prevalence of diabetes worldwide is estimated to be 9.3% (463 million people) in 2019. This prevalence is expected to rise to 10.2% (578 million) till year 2030 and will reach 10.9% (700 million) till year 2045.³

The International Diabetes Federation (IDF) has ranked Pakistan at 10 position out of 221 countries around the world, with 7.5 million cases of diabetes in age group 20 to 79 years.⁴ According to a study conducted in 2019 the prevalence of type 2 diabetes in Pakistan was 16.98%.⁵

Leptin is an "anorexigenic" hormone, mainly secreted by adipocytes. It influences the intake of food via its action on the hypothalamus by suppressing the appetite. As obesity causes leptin resistance it results in hyperleptinemia. There is a well-established link between elevated leptin levels and the development of insulin resistance and type 2 diabetes,⁶ in addition to obesity and hypertension. Leptin is also involved in stimulation of inflammatory reactions, atherogenesis, thrombosis and oxidative stress, leading to endothelial dysfunction and development of atherosclerotic plaques. Leptin is also associated with angiogenesis, bone homeostasis and reproduction.⁷

C-Reactive Protein (CRP), an acute phase protein is produced in the liver as a response to interleukin-6 (IL-6).⁸ CRP is a marker of inflammation and endothelial dysfunction and has a predictive role in cardiovascular disease. Insulin resistance and BMI have well established correlation with high sensitive C-reactive protein (hs-CRP) levels.⁹

It is suggested that CRP interferes with the insulin signaling pathway which results in insulin resistance.⁹ Despite a large number of studies, the role of hs-CRP in the pathogenesis of cardiovascular disease and type 2 diabetes remains controversial.⁹

Homocysteine (Hcy), formed after methionine demethylation, is a sulfur containing amino acid. Methionine being an essential amino acid is obtained by recycling it from diet. Hyperhomocysteinemia is a well-known risk factor for atherosclerosis.¹⁰ It can promote all the factors which lead to initiation of process of atherosclerosis including proliferation of vascular wall smooth muscle, and increasing oxidative stress.¹⁰ Plasma homocysteine levels are elevated in both pre-diabetics and type 2 diabetic patients due to the fact that insulin concentration influences plasma Hcy concentrations. The risk of cardiovascular diseases is 2-4 folds higher in diabetic individuals as compared to normal healthy subjects.¹¹ So the present study was conducted to evaluate the correlation between the adipocyte biomarker leptin, the cardiovascular biomarker hs-CRP, and homocysteine, in type 2 diabetes mellitus.

MATERIALS & METHODS

This cross sectional study was conducted in Hayatabad Medical Complex (HMC) Endocrinology ward Peshawar; laboratory analysis was done in Rehman Medical Institute (RMI) Peshawar. The study was conducted from January 01, 2018 to June 30, 2018 after obtaining ethical approval from the hospital board. The sample size based on WHO formula was calculated to be 96 with prevalence taken as 16.9%⁵ and accepted margin of error kept at 7.5%. Non probability convenience sampling was done by taking the inclusion criteria as Type 2 diabetics with age 45-65 years admitted in endocrinology ward of HMC who gave informed consent for participation in the study. Exclusion Criteria was pregnancy, breast feeding, acute infection or chronic inflammatory disease, liver dysfunction, renal disease, and dysthyroidism.

A detailed medical history and physical examination was conducted on the subjects to exclude other above-mentioned comorbid conditions and to record diabetic complications. A 5 ml fasting blood sample was taken by venipuncture and transferred to EDTA and vacutainer tubes. It was brought to RMI laboratory where it was centrifuged and stored at -20°C.

Plasma levels of CRP, Hcy, HbA1c and serum leptin were measured. CRP was measured by immunoturbidometric method, Hcy was measured by Chemiluminescent microparticle immunoassay and leptin was measured by ELISA using various laboratory kits according to the protocol.

Data about age, gender, BMI, leptin, HbA1c, Hcy and hs-CRP were recorded in SPSS 20. Data were analyzed to measure the frequency and percentages for categorical variables while mean and standard deviation were calculated for numerical variables. Pearson's correlation coefficients were determined for numerical variables i.e. Leptin, HbA1c, BMI, Hcy, and hs-CRP.

RESULTS

Out of 96 subjects, 23 were males and 73 were females. The mean age was 51.3 ± 8.8 years. The mean duration of diabetes was 7.0 ± 6 years. The mean BMI was 29.0 ± 5.0 . Mean systolic blood pressure was 131 ± 20 mmHg, mean diastolic blood pressure was 83 ± 11 mmHg.

Table 1: Mean values of general parameters (n=96)

Variables	Mean	SD
Age (years)	51.3	08.8
Duration of diabetes (years)	07.0	06.0
Body Mass Index	29.0	05.0
Systolic Blood Pressure (mm Hg)	131	20
Diastolic Blood Pressure (mm Hg)	83	11

Table 2 shows that the mean HbA1c level was 10.1 ± 2.4 %. The mean hs-CRP level was 04.7 ± 2.9 mg/L. The mean homocysteine level was 09.8 ± 4.8 μ mol/L. Mean serum leptin was 12.7 ± 5.2 ng/ml.

Table 2: Mean values of specific parameters (n=96)

Variables	Mean	SD
HbA1c (%)	10.1	02.4
hs-CRP levels (mg/L)	04.7	02.9
Homocysteine levels (μ mol/L)	09.8	04.8
Serum leptin levels (ng/ml)	12.7	05.2

Table 3 shows the correlation of leptin levels with various parameters. The correlation with BMI was positive and significant ($r=0.418$, $p=0.01$). The correlation with HbA1c was also significant and positive ($r=0.207$, $p=0.04$). The correlation with duration of diabetes was also significant and positive ($r=0.223$, $p=0.03$). The correlation with hs-CRP was positive but not significant ($r=0.249$, $p=0.467$). The correlation with homocysteine was also positive and not significant ($r=0.348$, $p=0.10$).

Table 3: Correlation of leptin with various parameters

Parameter	Correlation with Leptin levels (r)	p-value
BMI	0.418	0.01
HbA1c levels	0.207	0.04
Duration of diabetes	0.223	0.03
hs-CRP levels	0.249	0.467
Homocysteine levels	0.348	0.10

DISCUSSION

The present study, conducted on type 2 diabetics, reported a positive association of serum leptin levels with BMI and HbA1c. The association of serum leptin level and BMI has been studied extensively worldwide in several populations and majority has reported a positive correlation between these two parameters.¹²⁻¹⁴ A local Pakistani study showed a strong positive association between leptin and body mass index. In this study an increased serum leptin levels were noticed in obese subjects without type 2 diabetes mellitus.¹⁵ Another case-control study

conducted on obese diabetics and non-diabetics reported significantly increased serum leptin levels in obese diabetic as well as non-diabetic subjects as compared with non-obese diabetics and non-diabetic subjects. The study also reported that serum leptin levels were positively correlated with serum insulin, C-peptide levels and BMI. They concluded that Leptin levels are significantly increased in obesity and have a role in development of insulin resistance and type 2 diabetes mellitus.¹⁶

Leptin levels has been studied in different ethnic groups and neighboring populations by various studies. A study conducted in Western India also showed serum leptin levels to be positively correlated with body mass index.¹² A Turkish study also showed positive correlation between leptin and BMI.¹⁷

In another study the oxidative stress parameters like thiobarbituric acid-reacting substances (TBARS), superoxide anion (O₂⁻), superoxide dismutase (SOD) activity and total sulphhydryl groups were measured and compared in type 2 diabetics and normal subjects. The study reported increased concentration of leptin in obese, overweight and normal weight type 2 diabetes mellitus patients. A positive correlation between oxidative stress status parameters and leptin in obese patients was observed. They concluded that both increased oxidative stress and increased leptin levels result from obesity and they can lead to type 2 diabetes mellitus development.¹⁸

In the present study we found a positive association between inflammatory and cardiovascular markers CRP, Hcy, and serum leptin levels. The potential role of leptin in production of inflammatory cytokines is that adipose tissue being the source of circulating leptin also synthesizes IL-6, which induces CRP synthesis in the liver. Leptin directly induces production of IL-6, resulting in further increased production of hepatic CRP.¹⁹ Leptin has an obvious role in inflammation and high levels of leptin are positively associated with systemic markers of inflammation such as CRP.²⁰ The correlation of hs-CRP and leptin was reported to be positive in a study conducted by Wang et al.²¹ Another study conducted by Zhang et al²² also reported a positive correlation of same strength between hs-CRP and Leptin. Uslu et al²³ studied the association of serum leptin and all the study parameters in type 2 diabetics; they also found the same correlation strengths as reported by the present study. Thus, serum leptin levels can be used as a useful marker of cardiovascular risk and adiposity.

CONCLUSION

A positive association was observed between serum leptin levels and BMI; a positive association of cardiovascular risk markers with serum leptin levels was also noted. The findings therefore endorse serum leptin levels as a useful marker of cardiovascular risk and adiposity.

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