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Original Article

Internal Medicine Section

Association between Serum Leptin Levels in Obesity and Type 2 Diabetes Mellitus

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ABSTRACT

Introduction: It is hypothesised that leptin, the gene product of the obese gene plays an important role in regulating body weight and has a potential role as a treatment tool for obesity and Type 2 Diabetes Mellitus (T2DM). There is little documentation describing the relationship between serum leptin and the indices of obesity such as Body Mass Index (BMI) and T2DM.

Aim: To determine the relationship of serum leptin levels with BMI and type 2 DM patients.

Materials and Methods: This observational study was conducted in 140 patients with known diagnosis of type 2 DM with and without obesity and non-diabetic patient with BMI ≥25 kg/m^2. Patient's BMI and waist hip ratio were noted. Estimation of fasting serum leptin, fasting serum glucose, serum Total Cholesterol (TC), serum Triglycerides (TG), serum High Density Lipoprotein (HDL) cholesterol, serum Low-Density Lipoprotein (LDL) cholesterol and Very Low Density Lipoprotein (VLDL) cholesterol were done. Kruskal Wallis test and Mann-Whitney U test was used to compare median of continuous variables. Correlation analysis was done using Spearman's correlation technique.

Results: Median serum leptin level was significantly higher in patients who had DM and whose BMI was ≥25 kg/m^2. Fasting glucose, TC, LDL cholesterol, and VLDL cholesterol showed statistically significant and positive correlation, whereas HDL-cholesterol showed statistically significant and negative correlation with serum leptin levels.

Conclusion: Serum leptin levels were higher in type 2 DM and obese patients. Fasting glucose, TC, LDL cholesterol, and VLDL cholesterol showed positive correlation, whereas HDL-cholesterol showed negative correlation with serum leptin levels.

Keywords: Correlation, Fasting glucose, Serum triglycerides

INTRODUCTION

Diabetes Mellitus (DM) has been a leading cause of mortality and morbidity worldwide, T2DM constitutes 90% of population of diabetes load worldwide [1]. India has emerged as diabetes capital of the world [2]. The prevalence of DM in India is predicted to be around 62 million [3]. Increased insulin resistance and higher waist circumference with lower BMI are more found in Indian phenotype, also known as "Asian Indian phenotype". It makes Indians more prone to DM and its complications.

Serum leptin can be considered as a biological marker reflecting total body fat. Leptin controls appetite and glucose metabolism; and is primarily secreted by adipocytes. Circulating leptin levels are directly proportional to the total amount of fat in the body [4,5]. Leptin deficiency or resistance leads to uncontrolled food intake, obesity, and diabetes mellitus, atherosclerosis, hypertension, and coronary vascular disease [6,7]. There has been an increase in the obese population worldwide [8]. People with obesity have higher chances of developing non-communicable diseases such as T2DM, hypertension, and other cardiovascular diseases [9].

In spite of corresponding low BMI, Indians have a unique body structure characterised by increased abdominal fat deposition [10]. Measurement of serum leptin levels is cost intensive, and requires sophisticated equipment and thus not accessible for routine health care, whereas, BMI, Waist Circumference (WC), and Waist-Hip Ratio (WHR), can be easily measured. So, if these parameters are proven to be effective in predicting serum leptin level, it would be beneficial. There is little documentation describing the relationship between serum leptin and the indices of obesity, especially among Indian population. Serum leptin levels are reported to be high in diabetic patients [3,11,12] whereas, Haffner SM et al., reported that no significant difference in leptin levels in diabetic and non-diabetic people [13]. Serum leptin levels were higher in obese individuals [14-16]. The aim of the study was to determine the relationship between serum leptin levels, BMI and type 2 DM.

MATERIALS AND METHODS

This cross-sectional observational study was conducted between July 2017 and October 2018 at Poona Hospital and Research Centre, Pune, India. After approval from the scientific advisory committee and Institutional Ethics Committee (Ref. No- RECH/EC 2017-18/371), written informed consent was obtained from all the patients. One hundred forty patients aged ≥18 years of either sex with known diagnosis of T2 DM with and without obesity and non- diabetic patients with BMI ≥25 Kg/m^2 were included. Patients with known diagnosis of type-1 DM, hypothyroidism, chronic renal failure, nephrotic syndrome, familial hypercholesteremic syndromes, liver disease, anaemia and patients already on lipid lowering drugs were excluded from the study.

Patient's height, weight, BMI and WHR were noted. Estimation of fasting serum leptin, fasting serum glucose, serum TC, serum TG, serum HDL cholesterol, serum LDL cholesterol and VLDL cholesterol was done. Fasting blood samples from the participants were collected by venipuncture in plain and ethylene diamine tetra acetic acid vacutainers. Serum glucose, TC, TG, HDL cholesterol, and LDL cholesterol were estimated using commercially available kits. TC, HDL cholesterol, LDL cholesterol, TG were tested by Cholesterol Oxidase Peoxidase method, direct measure polymer-polyanion, direct measure Homogenous method, and GPO-POD respectively. Serum leptin was measured by sandwich type enzyme linked immunoassay using commercially available Kit (Diagnostic biochem Canada Inc.) The data obtained was entered in a pre-tested study proforma and analysed.

Diabetes mellitus was defined if fasting plasma glucose was >126 mg/dL or 2 hours post-prandial >200 mg/dL or HbA1c >6.5 or

patient was on anti-diabetic medicine [17]. The WHO protocol for Asian population was considered for categorization of BMI [18].

On the basis of a previously published study [19], a sample size of 140 patients was calculated by a formula [20] with 80 % power and 5% probability of Type I error to reject null hypothesis.

STATISTICAL ANALYSIS

Data collected were entered in Excel 2007 and analysis of data was done using Statistical Package for Social Sciences (SPSS) version 21, IBM Corporation, USA. The data on categorical variables is shown as n (% of cases) and data on continuous variables is shown as mean±Standard Deviation (SD). The statistical significance of inter-group difference between medians was tested using Mann-Whitney U test. For more than two groups, Kruskal-Wallis H test was used. Correlation analysis was done using Spearman's correlation technique. The confidence limit for significance was fixed at 95% level with p-value <0.05.

RESULTS

As depicted in [Table/Fig-1], majority of the patients were between the age group of 40-<60 years. The mean age of the patients was 51.4 years. Majority of the patients had BMI ≥30.00 kg/m^2. The mean BMI of the patients was 30.16 kg/m^2. Majority of the patients had DM.

Baseline characteristics	N (%)
Age group in years	
<40	26 (18.6%)
40-<60	83 (59.3%)
≥60	31 (22.1%)
Mean age in years ±SD	51.4±11.1
Gender	
Males	74 (52.9%)
Females	66 (47.1%)
BMI in kg/m^2	
18.50<25	20 (14.3%)
25.00<30	45 (32.1%)
≥30.00	75 (53.6%)
Mean BMI in kg/ m^2±SD	30.16±5.05
Diabetes mellitus	
Yes	101 (72.1%)
No	39 (27.9%)

[Table/Fig-1]: Comparison of median serum leptin levels in diabetes mellitus and non diabetes mellitus patients.
SD: Standard deviation; BMI: Body mass index

The distribution of median serum leptin level was significantly higher in patients who had DM compared to patients without DM [Table/Fig-2]. Median serum leptin levels were significantly higher in patients whose BMI was ≥25 kg/m^2 [Table/Fig-3]. Fasting glucose, TC, LDL-cholesterol, and VLDL cholesterol showed statistically significant and positive correlation, HDL-cholesterol showed statistically significant and negative correlation whereas serum TG did not show statistically significant correlation with serum leptin levels [Table/Fig-4].

	Diabetes mellitus		
	Yes (n=101)	No (n=39)	p-value
Median serum leptin levels in ng/mL	71.5	31.5	0.001

[Table/Fig-2]: Comparison of median serum leptin levels in diabetes mellitus and non-diabetes mellitus patients.
Mann-Whitney U test was used

	BMI (kg/m^2)			
	18.50 <25.00 (n=20)	25.00 <30.00 (n=45)	≥30.00 (n=75)	p-value
Median serum. leptin levels in ng/mL	45.2	69.5	67.5	0.001

[Table/Fig-3]: Comparison of median serum leptin levels according to BMI levels.
Kruskal-Wallis H test was used; BMI: Body mass index

Correlation of serum leptin with	r-value	p-value
Age	0.470	0.001
BMI	0.121	0.160
WHR	0.145	0.084
Fasting glucose	0.549	0.001
Total cholesterol	0.268	0.002
Serum triglycerides	0.114	0.188
HDL-cholesterol	-0.314	0.001
LDL-cholesterol	0.269	0.001
VLDL-cholesterol	0.308	0.001

[Table/Fig-4]: Correlation of serum leptin levels with various parameters.
Spearman's correlation analysis was done; BMI: Body mass index; WHR: Waist hip ratio; HDL: High density lipoprotein; LDL: Low density lipoprotein; VLDL: Very low density lipoprotein

DISCUSSION

In the present study, median serum leptin levels were significantly higher in patients with DM compared to individuals without DM. Diwan AG et al., conducted a study in India reported that serum leptin levels were significantly high in non-obese diabetic patients (7.09±1.83 ng/mL) as compared to normal healthy controls (5.38±2.20 ng/mL). They further observed that mean serum leptin was significantly higher in obese diabetes patients (9.83±1.72 ng/mL) than normal controls (5.38±2.20 ng/mL) [3]. Tatti P et al., conducted a study in Italy, reported that higher serum leptin was recorded in DM patients, compared to non-DM individuals [11]. McNeely MJ et al., showed that among Japanese Americans, increased baseline leptin levels were associated with increased risk of developing T2DM in men but not in women. In accordance with these studies, incidence of high serum leptin was higher in DM patients than non-DM individuals [12]. McNeely MJ et al., conducted a prospective cross-sectional observational study, in which they followed the patients for 5-6 years. Haffner SM et al., demonstrated in Mexican-Americans that the leptin concentrations were not different in diabetic and non-diabetic and the association of leptin with the components of obesity, principally the BMI, was similar in diabetic and non-diabetic subjects [12,13].

In the present study, median serum leptin levels were significantly higher in patients whose BMI was ≥25 kg/m^2. Maffei M et al., who conducted study in USA reported that plasma leptin levels were highly correlated with BMI [14]. Al-Maskari MY et al., studied Omanis, obese and non-obese healthy participants and found that there was a significant difference in serum leptin levels between the two groups (34.78±13.96 ng/mL vs 10.6±4.2 ng/mL) and a significant positive correlation between leptin levels in obese participants with weight (p=0.002), body fat percentage (p=0.0001) and BMI (p=0.001) [15]. In present study, there was statistically significant correlation of serum leptin levels with BMI (r=0.121, p=0.160). Lele RD et al., conducted study in Mumbai, India. They reported a significant positive correlation of serum leptin levels with BMI and waist circumference among diabetic and healthy controls (p<0.001) [16]. In present study, there was no statistically significant correlation between serum leptin levels and WHR (r=0.145 and p=0.084).

Isidori AM et al., reported that serum leptin levels gradually decline during aging in Italian population and leptin reduction is higher in women than in men, (r=-0.25 in women and -0.23 in men, p<0.01 in obese and -0.22 in women and -0.20 in men, p<0.05 in non-obese) in adult humans of different body weight [21]. In this study, there was positive and statistically significant correlation between serum leptin levels and age (r=0.470, p=0.001). Van Den Saffele JK et al., proved that serum leptin levels, whether or not adjusted for BMI, were found to increase with age, the values tending to

level off after the age of 45 years, and were strongly correlated to BMI (r=0.77) and fat mass (r=0.81) assessed by the bio-impedance method in Belgians [22]. Linear regression analysis showed a similar slope for the relationship between BMI and serum leptin levels in the three age groups. In the present study, age, BMI and WHR did not show statistically significant correlation with serum leptin levels. Considine RV et al., who conducted a study in USA which stated that serum leptin levels can serve as an indicator of fat content and that its levels increases exponentially with increasing body fat percentage [23].

In the present study, TC (r=0.268, p=0.002), LDL-cholesterol (r=0.269, p=0.001), and VLDL cholesterol (r=0.308, p=0.001) showed statistically significant and positive correlation with serum leptin levels whereas HDL-cholesterol (r=-0.314, p=0.001) showed statistically significant and negative correlation with serum leptin levels. Serum TG levels did not show statistically significant correlation with serum leptin levels. Rainwater DL et al., in USA reported a significant positive correlation between serum leptin and HDL cholesterol and TG [24]. Leyva F et al., in United Kingdom reported a significant positive correlation between serum leptin with BMI (r=0.57, p<0.001), TG (r=0.31, p=0.007), fasting glucose (r=0.32, p=0.003), but no significant correlations emerged between plasma leptin concentrations and age, HDL cholesterol [25]. Brennan AM et al found an association between (raised) serum leptin and adipocyte dysfunction in presence of increased BMI, C reactive protein, LDL Cholesterol and TG [26].

LIMITATION

This was a cross-sectional study, which can simply bring an association but cannot predict the effect of individual variables. Multivariate analysis were not done. Prospective studies should be carried out to assess whether serum leptin levels can be used to predict the development of T2DM and obesity in the population. The sample size of the present research was small; hence multi-centric studies with large sample size should be conducted to validate the results.

CONCLUSION

Median serum leptin level was significantly higher in patients who had DM and whose BMI was ≥25 kg/m^2. Fasting glucose, TC, LDL cholesterol, and VLDL cholesterol showed statistically significant and positive correlation, whereas HDL cholesterol showed statistically significant and negative correlation with serum leptin levels.

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