

Relation Between Leptin and Insulin In Patients With Type II Diabetes Mellitus

Mohiti J, Afkhami M, Babaei A.

Yazd Diabetes Research Center, Shahid Sadoughi University of Medical Science, Yazd, Iran.

Since leptin and insulin are reported to control glucose metabolism, a relationship between these two hormones should reveal the metabolic effect of them hormones on glucose metabolism. In this study, the relationship between leptin and insulin was evaluated in obese type II diabetic patients, BMI>30 kg/m² (group A), and non-obese type II diabetic patients, BMI<25kg/ m² (Group B).

Materials and Methods: 49 subjects were studied. Of these, 32 subjects (4 male and 28 female) were group A and 17 subjects (8 male and 9 female) were group B; both groups were analyzed for Leptin, Insulin, and HbA_{1c}.

Results: The results obtained showed leptin, insulin and HbA_{1c} levels of 5.16±1.2 µg/L, 6.75±1.2 µIu/mL and 9.38±0.56% for group B, these levels being 21.8±11.2 µg/L, 11.44±5.8 µIu/ml and 8.76±0.36% for group A.

Conclusion: The results of this study show that plasma leptin concentrations increased with body mass index. Plasma leptin levels in obese diabetics, in comparison to non-obese diabetic individuals, were four times higher (p=0.001). Statistical analysis indicates a direct correlation between fasting blood leptin and insulin (r=0.598 p=0.05) in group B, while the opposite is seen in group A.

Key Words: Leptin, Insulin, Diabetes, Obesity, resistance, HbA_{1c}

Introduction

Leptin, a 16 kDa circulating hormone produced and released primarily by adipose tissue, exerts a regulatory control mechanism on food intake via inhibition of neuropeptide Y and increases the basal metabolism rate with selectively promoting fat metabolism.¹ Leptin has two types of receptors; the long form and humorous short form. At the beginning direct leptin actions were thought to be exclusively confined to the central nervous system (CNS). It is now clear that there are multiplicities of peripheral target organs such as the pancreas, skeletal muscle, liver and gastrointestinal system.^{2,3}

Leptin appears to play a range of roles as a growth factor in a number of different cell types, such as a mediator of energy expenditure and most importantly interaction with other hormonal mediators and regulators of energy and metabolism such as insulin, glucagon, growth hormone and glucocorticoids.^{5,6,7}

A large body of evidence indicates that leptin along with insulin exerts an inhibitory effect on food intake, and an activation effect

Correspondence: Javad Mohiti, Yazd Diabetes Research Center, Joomhori Avenue, Yazd, Iran.

E-mail: mohiti_99@yahoo.com

on the regulation of thermogenesis within the central nervous system.^{4,14} Leptin and insulin function as a critical signal to the brain in the long-term regulation of energy homeostasis.^{14,16} The exact relationship between leptin and insulin is not clear and is sometimes controversial.^{8,9} Some researchers have failed to show a direct effect of leptin on energy homeostasis,¹⁰ while other studies have focused on the relationship between leptin and insulin, which share many properties as adiposity signals. Although insulin is secreted from the pancreatic beta cells rather than from adipocytes, the secretion of both hormones is influenced by the overall amount of fat stores as well as by short-term changes in energy balance.¹⁴ Moreover, insulin receptors are located in the same key hypothalamic areas as leptin receptors, whereas insulin secretion is stimulated acutely in response to meals, leptin secretion is not.

Although the mechanisms governing leptin secretion have yet to be fully elucidated, insulin appears to play a key role. Most obese mammals have elevated plasma concentrations of leptin and insulin, and they appear to be resistant to leptin-induced anorexia.¹² Therefore, the relationship between these two hormones should be revealed through the metabolic effects of these hormones on energy balance. In this study, the relationship between leptin and insulin in obese and non-obese type II diabetic patients was evaluated.

Materials and Methods

This cross-sectional study was carried out on patients with type II diabetes mellitus who had been referred to the Diabetes Research Center of Yazd University of Medical Sciences. They were new cases of diabetes who were on a diabetic diet, receiving no insulin treatment and showed no significant diabetes complications. The diagnosis of diabetes mellitus was made according to the World Health Organization Expert Committee on

diabetes mellitus, Geneva: WHO 1985. Technical Report Series. 727.

Study design: 49 subjects were studied, of which 32 [(4 males and 28 females, mean age=54±9yr, BMI=36.6±3.7kg/m² (range, 31-47)] were type II diabetics (group A) and 17 [(8 males and 9 females, mean age=47±10yr, BMI=24.2±2.8 kg/m² (range 18-25) were type II diabetics (group B)].

After measuring weight and height, and obtaining other information needed, a blood sample was taken from each individual after 10-12 hours fasting and serum was obtained. Samples were immediately frozen at -70°C until the time of analysis.

Assays

Leptin analysis: Samples were thawed at room temperature and serum leptin concentrations were determined using a sensitive ELISA kit. DRG.Com. Germany This assay has a detection limit of 0.05 µg/L. Day to day CVs were 13% at 0.32 µg/L and 5.8% at 2.14 µg/L. Leptin ELISA.

Insulin analysis: Insulin concentrations were detected from serum using a human ELISA test kit (Q-1-DIAPLUS, USA) after the serum samples were thawed at room temperature. This assay has a sensitivity margin of 0.5 µIU/ml.

HemoglobinA1c measurement: HbA1c was measured by Ion-Exchange chromatography using the DS5 Pink-300 test kit (Drew Scientific Limited. UK) from whole blood immediately after obtaining the sample.

Body mass index (BMI) was calculated for each case as weight (in kilograms) divided by height (in meters) squared. Glucose, triglycerides and cholesterol were measured by a colorimetric method using Autoanalyser RA-1000. Other kits and substances were obtained from Sigma chemical company.

Table 1. The characteristic and results analysis of obese and non-obese Type II diabetic patients

Variable	Group A (n=32)	Group B (n=17)	P
Leptin (µg/L)	21.8±11.2	5.16±1.20	0.001
Insulin (µIu/mL)	11.44±5.8	6.75±1.20	0.078
FBS (mg/dL)	174.62±9.97	177.58±13.25	0.86
HbA1c (%)	8.76±0.36	9.38±0.56	0.340
Cholesterol (mg/dL)	222.15±11.82	194.05±9.38	0.053
Triglycerides (mg/dL)	219±11.18	254±41.10	0.299

Statistical analysis

Data were analyzed using the SPSS statistical package program. The obese group was compared to the non-obese group using two-tailed students' t-test. Correlation between the groups was tested by the Pearson test. A P value <0.05 was considered statistically significant.

Results

As shown in table 1, the mean level of leptin obtained was 21.8±11.2 µg/L in group A and 5.16±1.2µg/L in group B. These results show that plasma leptin concentrations in group A increase with body mass index. Plasma leptin levels in obese diabetics (group A) when compared to those of non-obese diabetic patients (group B) show more than a four-fold increase (p=0.001, table 1).

Our statistical analysis results show a significantly positive correlation between leptin and insulin (r=0.59, p=<0.05) in group B (table 3), the correlation however in group A (table 2) between leptin and insulin is negative and not significant (r=-0.089, p=0.62).

The mean insulin levels obtained were 11.4±5.8 µIu/ml in group A and 6.75±1.2 µIu/ml in group B (table 1), The mean values of HbA1c obtained were 8.76 %± 0.36 in

Table 2. Correlation between Leptin and Insulin, FBS, HbA1c, Cholesterol, Triglyceride (TG), and BMI in group A

Leptin	r	P
Insulin	-0.089	0.62
FBS	-0.213	0.242
HbA1c	-0.095	0.605
Cholesterol	0.050	0.786
TG	-0.101	0.582
BMI	0.211	0.245

Table 3. Correlation between Leptin and Insulin, FBS, HbA1c, Cholesterol, Triglyceride (TG), and BMI in group B

Leptin	r	p
Insulin	0.598	0.05
FBS	-0.190	0.464
HbA1c	-0.273	0.289
Cholesterol	0.361	0.154
TG	-0.032	0.903
BMI	0.179	0.327

results of the measurements obtained for FBS, triglycerides and cholesterol of both groups are shown in table 1.

Discussion

The paradoxical observation contributes to the question of whether alteration in leptin action contributes to diabetes caused by obesity or lipodystrophy merely as a correlate with these phenomena or if they occur as a consequence of insulin resistance.^{12,13} The

statistical analysis shows a significantly positive correlation between leptin and insulin in group B (table 3).

These correlations may express a cooperative effect between these two hormones in the control of body weight in group B,¹⁴ while the correlation between leptin and insulin is negative and not significant in group A (table 2) that may express an uncooperative effect of these two hormones on the control of body weight in this group.^{18,19} Our results indicate that plasma insulin levels are higher in either obese diabetic patients or obese non-diabetes individuals (results not shown) when compared to non-obese diabetic patients (table 1). Since other factors measured are very similar in the two groups (see table 1), the increase in insulin levels in group A may be caused by the impaired action of leptin signaling in cells.^{20,21} Kellerer and co-workers²¹ have assessed the effects of insulin concentration on the leptin signaling pathway in rat-1 and HEK293 cells. Their results suggest that insulin concentration may contribute to the pathogenesis of leptin.^{21,22} It has also been reported that leptin effects on glucose metabolism differ between lean mice and hyper-

glycemia and hyper insulinemic obese animals.^{19,23}

The observation that leptin levels are elevated in proportion to body fat is a generally accepted idea i.e. most obese individuals are leptin-resistant.^{20,21} Resistance to the actions of leptin could be caused by decreased leptin transport through the blood-brain barrier^{25, 26} or to reduced signaling, distal to the leptin receptor.^{24,25} Peripheral signals such as glucocorticoids may also interfere with leptin's interaction with its receptor and produce central leptin resistance.^{27,28}

Conclusion

The similarity of fasting levels of plasma HbA_{1c}, glucose and triglycerides seen in both the obese and non-obese groups lead us to conclude that circulating leptin and insulin levels appear to be the best biological markers of obesity that are related to insulin resistance in the obesity syndrome.

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