Insulin effect on Leptin Concentration in Children with New Onset Insulin Dependent Diabetes Mellitus

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Abstract

Objective: Serum Leptin concentration reflects the body fat mass. There is controversial reports about the insulin effect on serum Leptin concentration. We wanted to examine the effect of insulin therapy on serum Leptin in children with new onset type I diabetes.

Materials and Methods: This was a Cross-Sectional study in Children's Medical Center on 34 children who had new onset type I diabetes. Serum Leptin level was measured at presentation, as a baseline before initiation of insulin therapy, three to five days and after three months of beginning of insulin therapy. The linear regression by SPSSv.16 used for analysis.

Results: There was a meaningful difference between the baseline level of Leptin and 3th-5th day. (P=0.00). The dosage of insulin was the most important factor affecting the Leptin levels after three months of treatment (P=0.006). In third months of follow up, sex and insulin dosage were the variables that effected on Leptin level. (P=0.003).

Conclusion: The results of our study showed that children with new-onset type I diabetes have low Leptin level before insulin therapy which increased in 3-5 days after treatment. We concluded that the acute insulin therapy alters the Leptin secretion/ action.

Keywords: Leptin, Insulin, Type I diabetes, Children.

Introduction

Leptin is a 16 Kd protein with 167 amino acid that synthesized by adipose tissue and its gene is mutated in ob/ob mice. (1) Obese gene (ob) regulates energy balance in the mouse and human. Loss of function mutation causes morbid obesity in mouse and human. (2) Leptin may act at the arcuate nucleus of hypothalamus and reduces the production of neuropeptide Y (NPY), one of the most potent stimulators of food intake (3,4,5). There is a relationship between Leptin level, dementia and Alzheimer disease in asymptomatic old people (6). It can also regulate bone mass through neuroendocrine pathway. (7,8)

There are several reports suggesting Leptin as regulator of insulin (9,10), but insulin level is the same in normal people and type 2 diabetic patients .(10) Therefore chronic high endogenous insulin level does not cause

increased serum leptin concentration, but insulin and glucose infusion for two days increase it .(12)

Diabetic children are prone to growth retardation, delay puberty and obesity even they treated with new types of insulin .One study revealed circulating Leptin concentrations correlated with body mass index (BMI) and calorie intake per kilogram per day. It also showed that over substitution by insulin and increased food intake stimulated fat synthesis, increasing BMI, and subsequently induces Leptin secretion. According this study higher Leptin level in poor control diabetic children with higher glycosylated hemoglobin (HbA1C) concentration was explained by increased appetite and BMI. (13)

The insulin regulates ob gene expression in rat, regardless to its glucose lowering effects. (14)

One search showed that two hours after subcutaneous injection of insulin, it is a signal for Leptin secretion from adipose tissue of diabetic rats, maybe by transferring glucose into the fat cells. (15)

Poor glycemic control in diabetic patients may lead to increase serum levels of sOB-R (soluble leptin receptor) that is independent of Leptin secretion, but may have an impact on Leptin action .Excess of sOB-R related to Leptin could reduce Leptin sensitivity. (16)

There are some conflicting reports that didn't show any relationship between insulin and Leptin concentration. (17) Therefore this study conducted to examine the probable connection between serum Leptin level and insulin therapy in type 1 diabetic patients.

Materials and Methods

This cross sectional study carried out in Children's Medical Center, Tehran, Iran, Tehran University of medical sciences from June 2009 to May 2011.

We consecutively enrolled 34 new onset type one diabetic children and adolescents referred to emergency department or outpatient clinic of Children Medical Center.

Inclusion criteria were: polydipsia and polyuria and random blood glucose higher than 200 mg/dl, or fasting blood sugar higher than 125 mg/dl. Exclusion criteria were: severe obesity, chronic renal, rheumatic, hepatic, pulmonary and neurologic diseases.

We classified our patients into two groups including diabetic ketoacidosis and non-ketoacidosis according signs and symptoms and arterial blood gases (ABG).

At first, before insulin consumption, weight and height were measured and BMI was calculated using formula: [weight (kg)/height^2 (m^2)] and appropriate work up were performed for each subject. Before starting insulin therapy, Leptin level for each patient was measured.

We reassessed weight, height, BMI and also withdrew blood of subjects after 3-5 days and 3 months of insulin therapy again.

At third month of treatment glycosylated hemoglobin level (HbA1C) was measured and mean insulin dosages was calculated.

Patient´s withdrawn blood samples were centrifuged to extract serum and then it was frozen at -20 ºC.

Serum Leptin level was measured using DRG Leptin (sandwich) ELISA method.

Calculated data was analyzed by SPSS software version 16 with considering level of significance <0.05. At first descriptive analysis was presented for the data, then mean and standard deviation were calculated. The relationships between variables were investigated by paired t test and multivariate linear regression analysis

Ethical issues: This study was approved by Ethics Council, Tehran University of Medical Science. All the Helsinki criteria were considered in this study .We recorded all results without name and entered them in the database information by code name We included only patients whose parents agreed to take part in the study by verbal consent.

Results

Thirty four children (17 female and 17 male) with mean age of 6.07±2.46 years (between

2.6-11.3 years) were included. The mean weight, height, BMI at the first, second and third visit are shown in table 1.

The mean weight, height and BMI of two genders were different but there were no statistically significant differences between them. (*P*:0.659, *P*:0.507, *P*:0.574)

The mean serum Leptin levels at the first, second and third assays were: 1.34±2.03; 1.81±2.19; 3.45±3.49 ng/ml consequently.

The mean serum Leptin levels difference at the first and second assays were statistically significant. (*P*<0.000) but unlike difference of first and third serum Leptin levels they are not statistically significant. (*P*:0.184)

The mean serum Leptin levels difference of second and third assays are statistically significant. (*P*:0.131)

The patients were divided to two groups according to be ketoacidotic (DKA) or non ketoacidotic (non DKA). Non DKA were 28 patients and DKA affected were 6 cases.

The mean serum Leptin levels of non-DKA patients at the first (1.32±2.19 ng/ml) and second(2.03±2.36 ng/ml) assays were higher than in DKA affected patients (0.26±0.26 ng/ml ,0.77±0.4 ng/ml consequently), but in third assay it was lower in non-DKA affected patients comparing with DKA cases (3.05±2.82 vs. 5.34±5.69).

But the mean serum Leptin levels difference are not statistically significant. (*P*:0.254, *P*:0.205, *P*:0.148)

The mean HbA1C at third assay was 8.17±0.62 .Multivariate linear regression analysis didn't show a significant relationship between the first serum Leptin level and HbA1C in third month.(*P*:0.196)

According multivariate linear regression analysis there aren't any relationship between age, gender, BMI, height ,weight and the first serum Leptin level. (*P*:0.605)

The only factor affecting the second serum Leptin level was the first serum Leptin level. (*P*:0.000)

The only variable affecting the third serum Leptin level was gender. (*P*:0.003) and others (age, 1st &2nd &3rd BMI, 1st &2nd serum Leptin level) didn't influence on the third assay.

The mean third serum Leptin level in male gender was: 2.06±1.87 ng/dl and in female was: 4.84±4.19 ng/dl.

The mean serum Leptin level at third assay had relation with the mean Insulin dosage and it was higher in patients received larger insulin dosage. (*P*:0.006; X^2=19.64).

Discussion

This research was performed to study the effect of insulin therapy on Leptin level in patients with new onset type one diabetes mellitus .Thirty four new onset type one diabetic children were participated in this investigation. The results displayed serum Leptin level was lower than normal in new onset type one diabetic children before insulin treatment, but it increased after 3-5 days and 3 months of insulin therapy. Our findings were according with regulatory role of insulin for Leptin secretion that was mentioned by majority of other investigators. (9,11,12)

Several animal studies demonstrated effects of exogenous insulin on the serum Leptin level in rats. (12,14)

We showed that the serum Leptin level at 3-5

Table 1. Descriptive analysis weight, height, BMI

Variable	Mean	Standard deviation	Minimum	Maximum
Weight¹ (kg)	20.14	7.40	10.00	38.00
Height¹ (cm)	114.60	16.40	87.00	147.00
BMI¹ (kg/m²)	14.77	1.61	11.31	17.58
Weight² (kg)	21.05	7.50	10.5	39.5
BMI² (kg/m²)	15.48	1.57	11.8	18.20
Weight³ (kg)	22.64	7.77	11.0	41.00
Height³ (cm)	113.53	24.64	11.0	152.0
BMI³ (kg/m²)	16.21	1.67	12.5	18.67

1-the first assay
2-second assay
3-third assay

days of treatment was not affected by some variables such as height, weight, BMI, gender or age at first, before insulin treatment (*P*:0.06) but it was impressed by Leptin level before starting exogenous insulin (*P*:0.000) and it raised more significantly if the first serum leptin level was higher.

Although the serum level of Leptin was higher significantly at third month of treatment with insulin but it was not statistically significant.

The serum Leptin level of female patients was higher than male patients at third assay and its difference was statistically significant (*P*:0.03)

Our results resembled to other study that revealed upper serum Leptin level in girls. (9,10)

Unlike other investigations we didn't find any relationship between BMI and serum Leptin level (13) ,maybe due to our small sample size or lower BMI of patients in this study (mean male BMI:15.02±1.31 kg/m^2 ,mean female BMI:14.52±1.87 kg/m^2)

We demonstrated meaningful relationship between serum Leptin level and insulin dosage. As far as increment of insulin dosage, patients had upper serum Leptin level (*P*:0.006)

There are several survey findings consistent with our study that found insulin as a strong stimulator to secret Leptin in new onset diabetic patients. (9,10,11,12)

In vitro studies on rat adipocytes revealed glucose and insulin metabolism relationship with serum Leptin level. (14)

In patients with DKA the serum Leptin level was lower than non-DKA at first and second assays but it escalated to higher level than in non-DKA at third month of treatment (figures1-4). Perhaps due to higher dosage of insulin need based on severity of its deficiency.

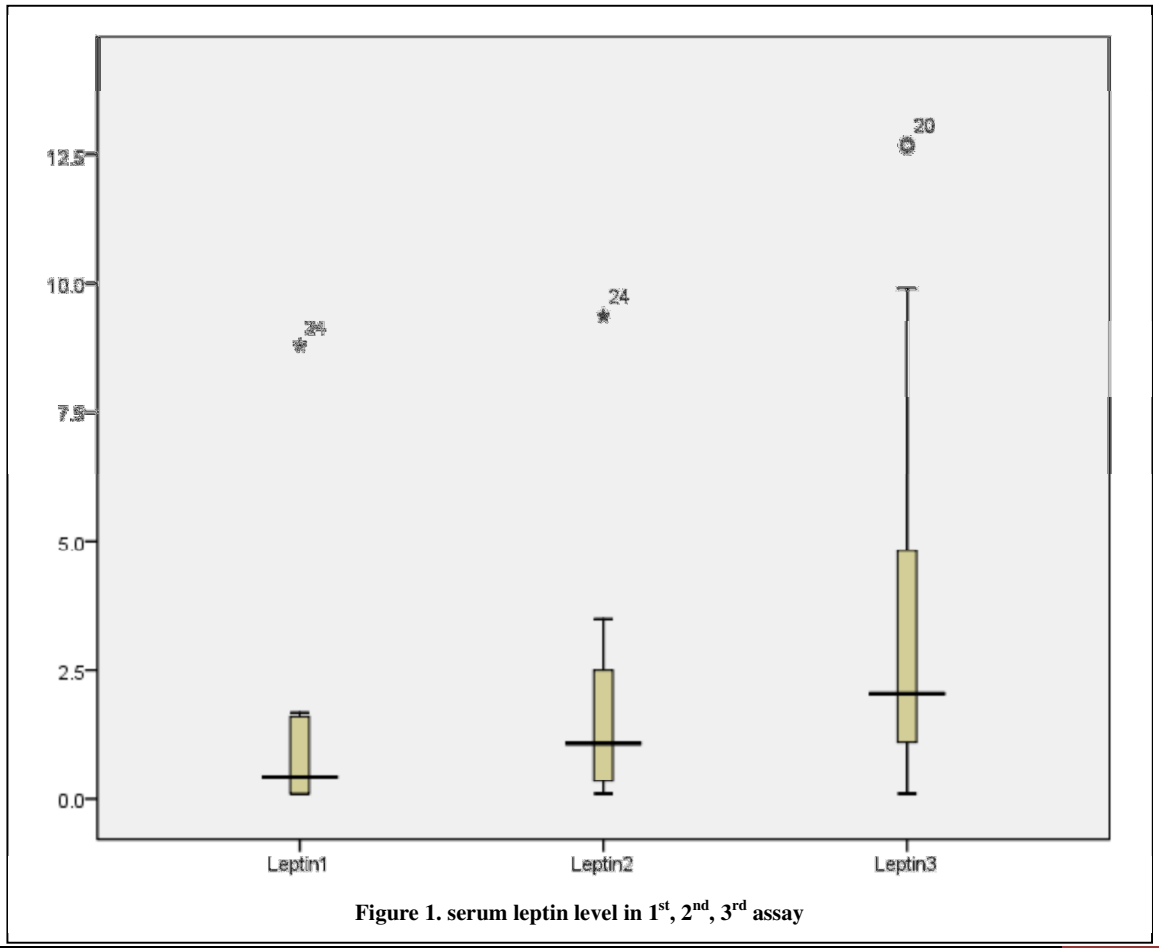

Figure 1. serum leptin level in 1st, 2nd, 3rd assay

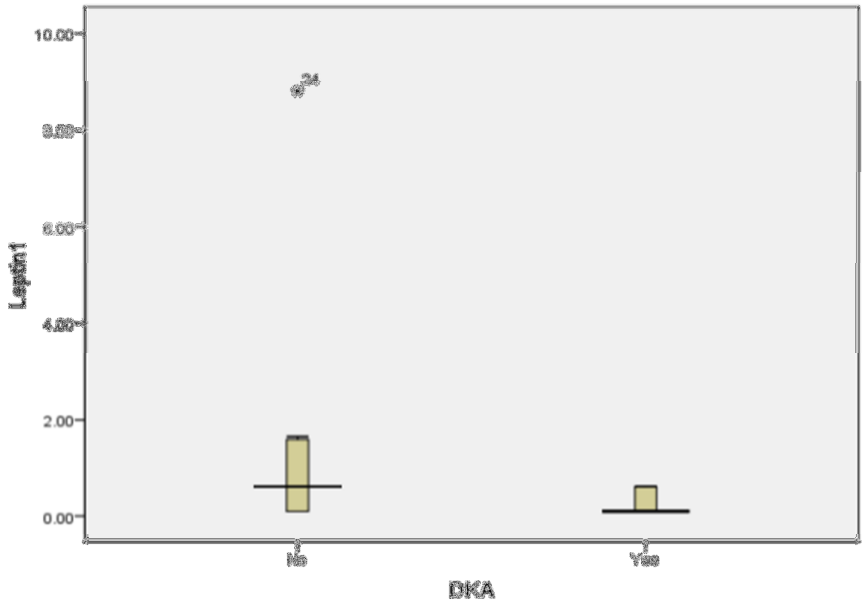

Figure 2. serum leptin level according DKA or non-DKA in 1st assay.

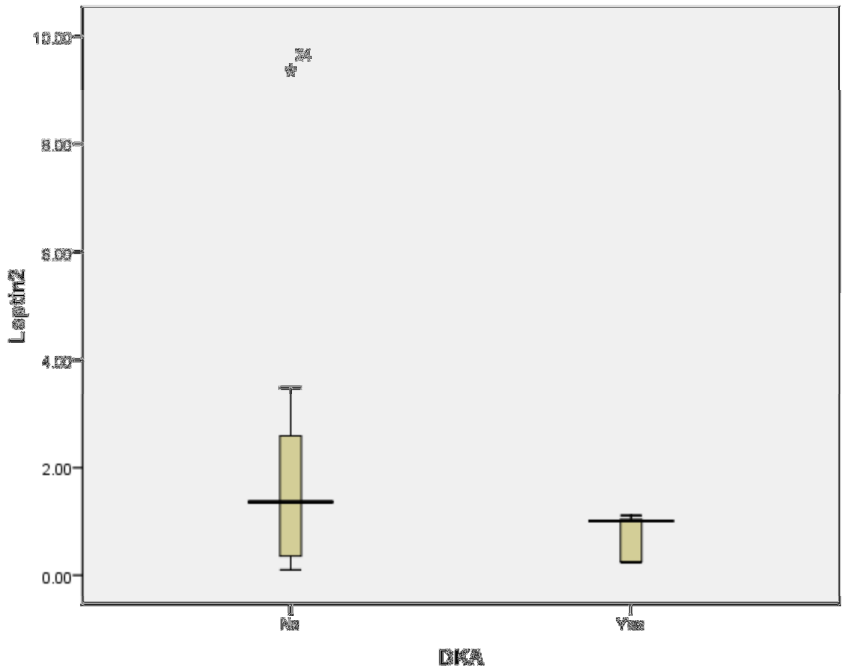

Figure 3. serum leptin level according DKA or nonDKA in 2nd assay

Conclusion

Regarding low insulin and Leptin level in new onset type one diabetic patients and significant increment of serum Leptin level after insulin treatment we conclude that acute insulin therapy could stimulate Leptin synthesis and

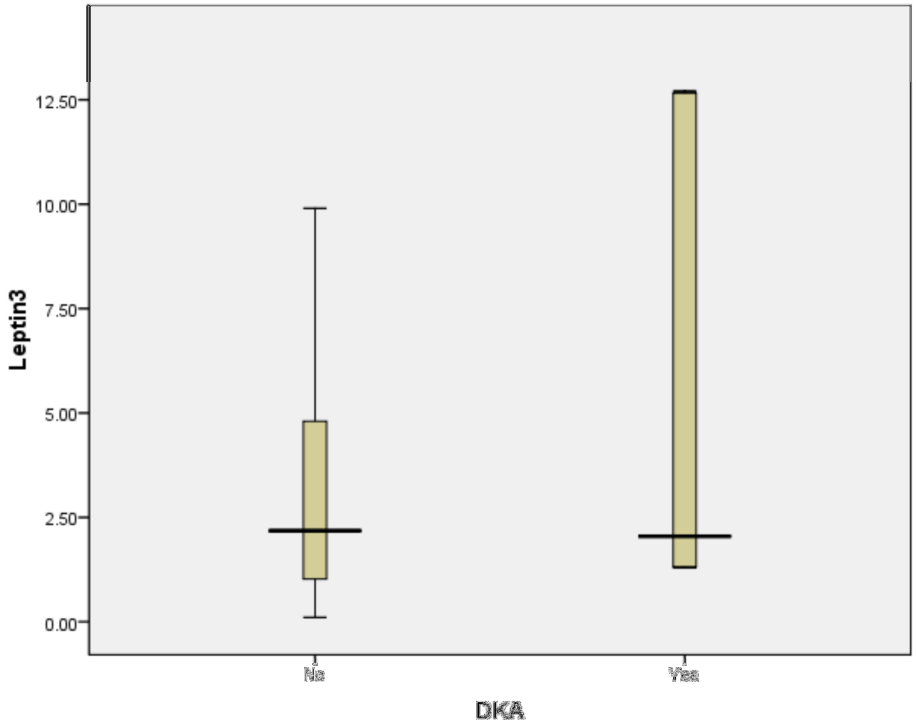

Figure 4. serum leptin level according DKA or non-DKA in 3rd assay

secretion. We recommended a multicenter study and comparison with control group. Trial therapy with Leptin and investigation of diabetic control and complications in treated group.

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