Plasma leptin levels in infants of diabetic mothers in fasting and satiety states

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SUMMARY. Cinaz P, Bideci A, Çamurdan MO, Şen E, Çamurdan AD. Plasma leptin levels in infants of diabetic mothers in fasting and satiety states. Turk J Pediatr 2004; 46: 142-146.

Infants born to diabetic mothers have elevated cord blood leptin levels. The aim of this study was to investigate whether the situation persists at the 2nd postnatal day, taking the fasting and satiety states into account and the influence of fetal exposure to hyperinsulinemia, which are proven important contributing factors to plasma leptin levels. Twenty infants born to mothers with gestational diabetes (Group I) and 20 controls (Group II) were included in the study. Groups were similar for sex and anthropometric measurements. Group I had higher leptin concentrations compared to Group II in fasting and satiety states ($p<0.01$). Fasting state leptin levels were significantly lower than seen in satiety in both groups ($p<0.01$). There was a positive and significant correlation between leptin concentrations and body mass index of infants. Leptin concentrations were positively correlated with plasma insulin levels in Group I. These findings suggest that plasma leptin levels are high in both fasting and satiety states on the 2nd postnatal day in infants born to mothers with gestational diabetes. The possible mechanism underlying this condition is fetal exposure to hyperinsulinemia due to hyperglycemia. The uniqueness of this report are that fasting and satiety states were taken into account and that the data was collected from the samples taken on the 2nd postnatal day, thus reflecting the exact milieu of the infant excluding the effects of the mother and the placenta.

Key words: infants of diabetic mother, plasma leptin, fasting, satiety.

Leptin, the protein encoded by the *ob* gene, is produced by adipocytes and is involved in regulation of body weight, fat content and energy balance via stimulating energy expenditure and suppressing appetite by working as an afferent signal from the peripheral fat mass to the hypothalamus[1-3].

Leptin in circulation is known to be affected especially by gender, hormonal status (especially insulin, sex steroids, glucocorticoids), and nutirition[4-6]. Along with these, there are several factors that influence plasma leptin levels such as the timing of the sample (i.e., day after delivery and the fasting/satiety states)[5-8].

The well known interaction of leptin with insulin and the possible effects of it on intrauterine growth have led researchers to investigate whether leptin levels change in infants born to diabetic mothers. At present, there are some what conflicting results related to this issue, most of them being studied in cord blood without taking fasting and satiety states into account[9-12].

In this study our aim was to investigate whether a difference exists in plasma leptin levels in infants born to mothers with gestational diabetes mellitus (GDM) and the influence of fetal exposure to hyperinsulinemia on leptin levels. Our study presented a new dimension by focusing on the satiety-fasting states and acquiring postnatal blood samples.

Material and Methods

The local ethics committee approved the study, and informed consent was taken from all the parents. Twenty newborns of mothers with GDM with gestational age of 39.6±1.5 weeks, birth weight of 3.93±0.82 kg, birth length of 52±3 cm and body mass index [BMI-defined as weight (kg) divided by the square of height (m^2)] of 14.45±2.08 kg/m^2 (Group I) and 20 sex,-gestational age,- birth weight-

matched newborns of non-diabetic mothers with gestational age of 39.2±1.2 weeks, birth weight of 3.45±0.30 kg, birth length of 50±1 cm and BMI of 12.65±0.42 kg/m^2 (Group II-the control group) were included in the study. Infants taking any medication, those with asphyxia fetal distress, major congenital anomalies, chromosomal disorders, or proven perinatal infection and those whose mothers received corticosteroids or other hormonal therapy were excluded. Five infants in Group I and three in Group II were born via cesarean section. Birth weight was assessed using a digital scale and then evaluated according to Lubchenco's intrauterine growth curve[13]. All mothers were on diet therapy for diabetes with non receiving insulin. All infants were exclusively breast-fed. Two blood samples were taken from infants on the morning of the 2nd postnatal day: a satiety sample at the 30th minute after feeding and a fasting sample taken at the 120th minute via a #23 butterfly venous puncture set. Samples were collected in ice-chilled plastic tubes containing 10 IU heparin/ml[14]. Plasma was obtained by centrifugation at 4°C for 10 minutes and then frozen at –70°C until the time of assay. Leptin, insulin and glucose levels were measured in all samples using a commercially available kit (Linco research IMC^R), radioimmunoassay (Diagnostic Products Corporation, LA), and glucose-oxidase method, respectively. Mothers' HbA1c levels were measured using spectrophotometry (ion exchange colon chromatography).

Statistical analysis comparing the two groups was made using the non-parametric Mann-Whitney U test. Correlation analysis was performed using Pearson's correlation method. Statistical significance was assumed at the conventional level of $p<0.05$. All analyses were two-tailed and performed with the SPSS software version 10.0 for Windows.

Results

There were no significant differences in sex, BMI, or absolute birth weight or height between the two groups.

Infants in Group I had significantly higher plasma leptin levels than infants in Group II in both fasting (22.97±10.05 ng/ml and 2.17±0.52 ng/ml, respectively; $p<0.01$) and satiety states (35.78±16.58 ng/ml and 3.34±1.14 ng/ml, respectively; $p<0.01$). Fasting state plasma leptin levels were significantly lower than the levels in satiety in both groups (p values <0.01) (Fig. 1).

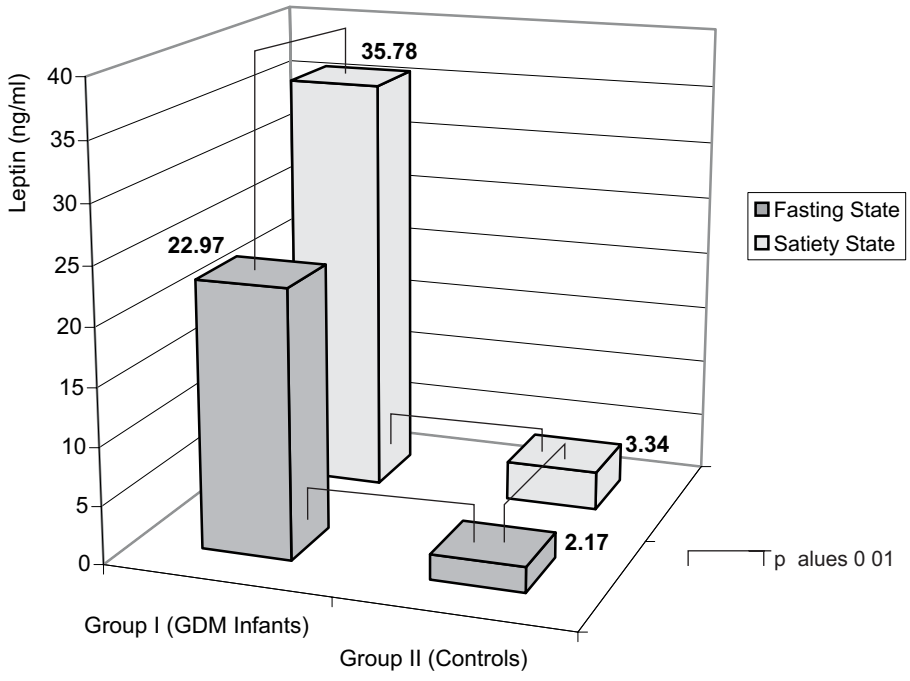

Fig. 1. Plasma leptin levels in both groups in fasting and satiety states. GDM: gestational diabetes mellitus.

There was a positive and significant correlation between plasma leptin levels and BMI of infants (Table I). Fasting and satiety plasma leptin levels were positively and significantly correlated with plasma insulin levels in Group I (Table II). All mothers were moderately controlled diabetic patients according the HbA1c levels, with a mean of 7.4±0.7%, and maternal plasma HbA1c levels were correlated with plasma leptin levels in both fasting and satiety states in Group I ($r=0.45$, $p<0.05$).

Table I. Corelation Between Plasma Leptin Levels with BMI in Both Groups

	Leptin (ng/ml)	BMI (kg/m^2)	r value	p value
Group I GDM Infants	22.97±10.05	14.45±2.08	0.46	<0.01
Group II (Controls)	2.17±0.52	12.65±0.42	0.53	<0.01

GDM: gestational diabetes mellitus; BMI: body mass index.

Table II. Corelation Between Leptin and Insulin Levels in Infants Born to Gestational Diabetic Mothers

	Leptin (ng/ml)	Insulin (µm/ml)	r value	p value
Fasting State	22.97±10.05	14.42±2.72	0.63	<0.01
Satiety State	35.78±16.58	18.65±4.72	0.49	<0.05

GDM: gestational diabetes mellitus; BMI: body mass index.

Discussion

Leptin is widely studied in adults and its role in obesity and energy balance is for the most part clear. On the other hand, the function of leptin in infants is still under research because it is not clearly understood whether circulating leptin levels are the cause or the consequence of many physiological conditions in infants.

Leptin level in cord blood is well correlated with birth weight and BMI in gestational or type I DM and normal pregnancies[9,10,12,15]. Although there are conflicting results[12], this condition is also supported by the studies performed on LGA, AGA, and SGA infants[5,15]. In our study, it was shown that plasma leptin levels are significantly correlated with BMI and birth weight, and this was concordant with the previous findings.

Prolonged exposure to insulin is needed in order for it to raise plasma leptin concentration in humans[16]. On the other hand, leptin acts as an insulin counter-regulatory hormone and inhibits insulin production[17]. In GDM, insulin-leptin interactions at the state of pregnancy and the extension of this interaction in an infant's physiology is a matter of consideration. It is shown that there is all significant positive correlation between insulin and leptin levels in infants born to mothers with GDM, but this correlation is lacking in infants born to mothers with type I DM[9]. Our data supports the findings that leptin and insulin are well correlated in infants born to mothers with GDM, and this correlation exists both in fasting and satiety states.

The diabetic pregnancy is a complex situation, in which profound changes in hormonal metabolism and adipose tissue take place. Two different organisms (the mother and the fetus) with complicated insulin-leptin-intrauterine growth interactions are in progress. Infants of diabetic mothers, especially of those who show poor metabolic control, have been characterized by macrosomia and increased subscutaneous tissue[18,19]. It can be postulated that plasma leptin should be higher in infants born to diabetic mothers because of the increased fat content and the chronic exposure to hyperinsulinemia, but in vivo studies show conflicting results. Two recent studies are examples, with Tapanainen et al.[9] showing that both insulin and leptin levels are higher in infants born to diabetic mothers compared to weight-matched controls, being higher in type I diabetics than gestational diabetics, and Ng et al.[10] demonstrating no significant difference in leptin levels between infants born to diabetic mothers and controls. Although some other studies in literature support the findings of Tapanainen et al.[9], it seems the topic has to be investigated further because no relationship between leptin and insulin levels are found in these studies[11,12]. Our study, showing positive and significant correlation between maternal

HbA1c and infants' plasma leptin levels, confirms that infants born to mothers with GDM have elevated plasma leptin concentrations and that diabetic control level is one of the predictors of this leptin concentraton. The high and significant correlation between insulin and leptin concentrations supports this hypothesis and suggests that the possible mechanism underlying high leptin values is fetal exposure to hyperinsulinism due to hyperglycemia. Because there was no difference in gender, birth weight or BMI between groups, we can postulate that this difference originates from the diabetic state only.

Other important factors in our study were that fasting/satiety states were taken into account and that the samples were taken at the 2nd postnatal day. To our knowledge, there is no other study in children in which plasma leptin levels were measured on the 2nd postnatal day considering fasting/satiety states. The postnatal day and the fasting/satiety states are important factors influencing the plasma leptin levels measured. Plasma leptin levels fall rapidly and significantly after birth, and this phenomenon takes place especially on the 1st postnatal day around the 16th hour and remains almost constant on the following days[7]. Therefore, it is obvious that cord blood sample does not reflect the child's real status, because it is clearly affected by leptin originating from the placenta and/or the mother. It is thus obligatory to take the sample after the 1st postnatal day in order to evaluate the child's real physiologic milieu. Fasting and satiety states are other important factors influencing leptin levels. Plasma leptin levels are reduced at fasting and increased after feeding[7,8]. Our study, reporting lower plasma leptin levels in the fasting compared to the satiety state in both groups, is in agreement with the literature. The mechanism underlying this condition is not clearly understood. Data suggest that the caloric intake is one of the important factors controlling leptin levels[20,21]. Bado et al.[22] reported that the stomach is a source of leptin and is responsible for the rapid increase in the concentration of leptin in the satiety state. It has also been shown that the expression of the *ob* gene is reduced after fasting in humans and rats[3,23].

In conclusion, our results confirm that plasma leptin levels measured at the 2nd postnatal day in both fasting and satiety states are higher in infants born to mothers with GDM compared to non-diabetics. The leptin levels were highly and significantly correlated with insulin and the mothers' HbA1c levels, suggesting that poor metabolic control of diabetes leads to higher leptin levels observed in the child. Combined with the fact that groups were similar according to their sex and BMI, it can be postulated that high leptin levels observed in infants of diabetic mothers is due to the diabetic milieu of the prenatal life. The authors of this study hope that it brings a new insight to the literature, first by evaluating samples taken at the 2nd postnatal day, when infants' leptin status is free from the effects of leptin production and other hormonal changes taking place in the mother and the placenta, and second, by taking fasting and satiety states into account, which are proven factors influencing plasma leptin levels.

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