Leptin levels and antihypertensive treatment in preeclampsia

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ABSTRACT

Introduction: This study was carried out to investigate changes in the plasma leptin concentrations during preeclampsia treatment and to determine whether antihypertensive treatments, aimed at decreasing leptin levels, would improve foetal outcomes.

Methods: A prospective study was undertaken in 57 pregnant women with preeclampsia (37 with mild and 20 with severe preeclampsia) and 46 normal pregnant women who were matched in maternal and gestational age and body mass index. The mild preeclampsia group was treated with alpha-methyldopa, while the severe preeclampsia group was treated with a combination of alpha-methyldopa and nifedipine.

Results: The severe preeclampsia group had significantly lower platelet counts, higher systolic and diastolic blood pressures and elevated serum uric acid concentrations. Pre-treatment plasma leptin levels were significantly increased in the severe preeclampsia group (range 18.3–49.5) compared to the the mild preeclampsia group (range 20.7–45.4) and normal controls (range 8.6–19.2). Post-treatment plasma leptin levels in both the mild and severe preeclampsia groups (range 10.2–23.5 and 11.3–24.4, respectively) were statistically similar to those of the control group (range 9.1–20.7). Estimated foetal weight, intrauterine growth retardation and demise were statistically similar in the three study groups.

Conclusion: Plasma leptin concentrations were found to be elevated in women diagnosed with severe preeclampsia. However, the exact mechanism underlying the increased plasma leptin levels in preeclampsia and the functional role of leptin in the development of hypertension need to be further clarified. Leptin has a promising future as a valuable marker to identity women with a high risk for preeclampsia.

Keywords: antihypertensive, leptin, placenta, preeclampsia, pregnancy

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INTRODUCTION

Preeclampsia is one of the most recognised clinical causes of high-risk pregnancies. Although preeclampsia affects about 1%–2% of pregnancies in some European countries, its prevalence can be up to 10%–15% in some South American and African countries. Preeclampsia is characterised by hypertension and arteriolar vasoconstriction, which decreases the uteroplacental perfusion and results in placental hypoxia. Long-lasting placental hypoxia can in turn lead to foetal growth retardation.[1]

Under the supervision of the obesity gene, the adipocytes and placental trophoblasts synthesise and secrete a protein called leptin. Leptin decreases the body weight by acting through its hypothalamic receptors and reducing the food intake. This protein may also act as a metabolic signal for the neuroendocrine and reproductive systems.[2-4]

Trophoblasts are responsible for the significantly increased plasma concentrations of leptin during the first two trimesters of normal pregnancies.[2,3] This marked increase in the synthesis of leptin is attributed to the prominent alterations of maternal weight, energy expenditure and hormonal status. However, the exact role of leptin in the pathogenesis of high-risk pregnancies is still undetermined.[5-7] A number of cross-sectional studies have found a significantly higher plasma concentration of leptin in preeclamptic pregnancies when compared with normal pregnancies.[8-14] However, the effects of antihypertensive treatment on the serum leptin levels of pregnant women with preeclampsia have not been thoroughly studied.[15]

The present study was carried out to confirm the reported increase in plasma leptin in preeclampsia and to investigate changes in the plasma leptin concentrations following the initiation of an antihypertensive regimen to treat preeclampsia. The present study also aimed to determine whether antihypertensive treatment, by decreasing leptin levels, would improve foetal outcomes.

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Table I. Clinical and demographic characteristics of subjects.

Variable	Mean ± SD			p-value
	Normal pregnancy (n = 46)	Mild PE (n = 37)	Severe PE (n = 20)	
Age (years)	25.3 ± 2.7	24.1 ± 1.9	22.6 ± 0.8	0.44
Parity	1.7 ± 0.3	0.8 ± 0.2	0.5 ± 0.1	0.57
Gestational age (weeks)	36.2 ± 3.4	34.3 ± 1.7	35.4 ± 2.3	0.35
Body mass index (kg/m^2)	23.7 ± 2.9	21.9 ± 1.8	24.6 ± 3.4	0.66
Systolic BP (mmHg)	119.1 ± 6.6	143.2 ± 7.8	155.4 ± 9.9	0.03*
Diastolic BP (mmHg)	74.5 ± 7.2	91.9 ± 8.3	100.7 ± 10.1	0.03*
Platelet count (×10^6/mm^3)	240.1 ± 38.2	233.7 ± 36.4	219.6 ± 27.3	0.17
Serum creatinine (mmol/l)	65.2 ± 6.4	70.7 ± 5.2	68.8 ± 7.5	0.69
Serum urate (mmol/l)	0.22 ± 0.04	0.34 ± 0.03	0.39 ± 0.07	0.04*

PE: preeclampsia; SD: standard deviation; BP: blood pressure
* $p < 0.05$ was considered statistically significant.

METHODS

The present study was approved by the Ethical Committee and Institutional Review Board of Dr Zekai Tahir Burak Women Health Research and Education Hospital, Turkey. Written informed consents were obtained from all participants.

This prospective study was undertaken in 46 normal pregnant women and 57 pregnant women diagnosed with preeclampsia. Preeclampsia was diagnosed and classified according to the criteria specified by the technical bulletin of the American College of Obstetricians and Gynecologists and the National High Blood Pressure Education Program Working Group Report on High Blood Pressure in Pregnancy.[1] Hypertension was described as an increase of 30 mmHg systolic or 15 mmHg diastolic blood pressure (BP) as compared to the values before 20 weeks of pregnancy, or an absolute BP > 140/90 mmHg after 20 weeks gestation if the earlier values were unknown. Proteinuria was defined as > 0.5 g of urinary protein excretion. The exclusion criteria were diabetes mellitus, chronic hypertension, a known history of peripheral vascular disease, a previous history of antihypertensive treatment and a body mass index (BMI) < 19 kg/m^2 or > 30 kg/m^2. Participants were categorised into three groups: control, mild preeclampsia and severe preeclampsia. Mild preeclampsia was defined as a BP ≥ 140/90 mm Hg and < 160/110 mm Hg, and proteinuria ≥ 0.3 g/day and < 2.0 g/day, whereas severe preeclampsia was defined as a BP ≥ 160/110 mm Hg and proteinuria ≥ 2.0 g/day. The three study groups were matched in maternal and gestational ages and BMI.

The mild preeclampsia group was treated with alpha-methyldopa (Aldomet®, Abdi İbrahim, İstanbul, Turkey), 250 mg tablet, three times a day, while the severe preeclampsia group was treated with a combination of alpha-methyldopa and nifedipine. A single dose of nifedipine (Nidilat®, Sanofi Synthelabo, İstanbul, Turkey), 10 mg capsule, was administered when the maternal BP exceeded 170/110 mmHg despite the regular alpha-methyldopa treatment.

Venous blood samples for leptin concentrations were withdrawn from all participants before and seven days after the initiation of treatment. Venous blood samples for leptin were collected into siliconised glass tubes containing Na_2EDTA (1 mg/mL), centrifuged immediately at 6420 g (4°C) and stored at –70°C until assayed. Blood samples were also obtained for full blood count, creatinine and uric acid concentrations.

Total circulating plasma leptin concentrations (ng/ml) were measured in duplicates using a double antibody radioimmunoassay (Linco Research Laboratories, St Louis, MO, USA). The assay employed polyclonal anti-human (rabbit) antibodies raised against recombinant human leptin. Standards and I-125 tracers were also made from recombinant human leptin. The average intra-assay coefficients of variation (CVs) were 2%–5%, and the inter-assay CVs were 4% for low values (2.1–3.9 ng ml^{-1}), and 1% for high values (16.4–24.6 ng ml^{-1}). Complete blood count (CBC) parameters were measured by an automated blood counter (Cell-Dyn 4000, Abbott Diagnostics, Santa Clara, CA, USA). Serum creatinine and uric acid concentrations were measured by an automatic chemical analyser, TBA 40FR (Toshiba, Tokyo, Japan).

Age, parity, BMI, gestational age, systolic and diastolic BPs, treatment regimens, pre- and post-treatment plasma leptin levels, CBC parameters and serum concentrations of uric acid and creatinine, the estimated foetal weight, the existence of intrauterine growth retardation (IUGR) and intrauterine demise of all participants were recorded. The data collected was analysed using the Statistical Package for Social Sciences version 11.0 (SPSS Inc, Chicago, IL, USA). The values were expressed as the mean ± standard deviation (SD) whenever appropriate. Statistical analysis was performed using the Student's *t*-test or the ANOVA,

Table II. Clinical outcome of subjects.

	Mean ± SD/ no. (%)			p-value
	Normal pregnancy (n = 46)	Mild PE (n = 37)	Severe PE (n = 20)	
Pre-treatment leptin (ng/ml)	16.3 ± 2.8	30.6 ± 3.5	31.7 ± 4.9	0.01*
Post-treatment leptin (ng/ml)	17.8 ± 2.6	18.3 ± 2.9	19.1 ± 3.6	0.81
Estimated foetal weight (g)	2656.4 ± 479.1	2813.1 ± 527.4	2774.5 ± 511.8	0.72
IUGR	6 (13.1)	5 (13.5)	3 (15.0)	0.76
Intrauterine demise	1 (2.2)	1 (2.7)	0 (0.0)	0.77

PE: preeclampsia; SD: standard deviation; IUGR: intrauterine growth retardation
* $p < 0.05$ was considered statistically significant.

with Fisher's least-significance difference test. For all comparisons, statistical significance was defined as $p < 0.05$.

RESULTS

The demographic and clinical characteristics of the subjects who took part in the present study are shown in Table I. As expected from the recruitment criteria, the severe preeclampsia group had significantly higher serum uric acid concentrations as well as systolic and diastolic BP values. The three study groups were statistically similar in age, parity, gestational age, BMI, platelet counts and serum creatinine levels.

Table II shows the plasma leptin concentrations of the subjects before and seven days after the pharmacological treatment. Pre-treatment plasma leptin levels were significantly increased in the severe preeclampsia group (range 18.3–49.5 ng/ml) compared to the mild preeclampsia group (range 20.7–45.4 ng/ml) and the normal controls (range 8.6–19.2 ng/ml). Post-treatment plasma leptin levels in both the mild and severe preeclampsia groups (range 10.2–23.5 ng/ml and 11.3–24.4 ng/ml, respectively) became statistically similar to those of the control group (range 9.1–20.7 ng/ml). Table II also demonstrates the foetal status of the three study groups. The women with a normal pregnancy, and those with mild and severe preeclampsia did not differ significantly in terms of estimated foetal weight, IUGR and foetal demise.

DISCUSSION

Leptin is described as a major placental protein which exhibits metabolic and physiological functions in a normal pregnancy.[2] However, clinical and experimental trials have not yet clarified the definite role of leptin in the pathophysiologic mechanisms of high-risk pregnancies.[2-4] The present study was designed to investigate the altered plasma leptin concentrations in preeclampsia and after the initiation of an antihypertensive regimen for preeclampsia treatment. It also aimed to determine whether antihypertensive treatment, by reducing leptin levels, would improve foetal outcomes.

Leptin was used to evaluate the effects of antihypertensive treatment as it was thought to reflect the foetal metabolic status, which is known to be strongly related to IUGR and foetal demise. Previous studies have demonstrated that plasma leptin concentrations were increased significantly during the third trimester of preeclamptic pregnancies in contrast to normal pregnancies.[8-14] Other studies have documented that plasma leptin levels were elevated even before preeclampsia had become clinically evident.[16,17] The present study also showed a significant increase in the plasma leptin levels of women with mild and severe preeclampsia, suggesting the exaggerated placental production as the cause.

Increased placental production of leptin might be considered to be a response to placental hypoxia caused by severe preeclampsia. This supports the idea that augmented plasma leptin levels reflect placental hypoperfusion and/or hypoxia in severe preeclampsia. Therefore, leptin may serve as a marker of preeclampsia, indicating the associated placental hypoxia.[8,10] On the other hand, elevated leptin levels may represent the adaptation mechanism of the foeto-placental system, which attempts to compensate for the impaired placental perfusion and to provide the metabolic needs of the foetus. Preeclampsia induces inflammatory mediators, such as tumor necrosis factor-α and interleukin-6, which may in turn trigger leptin release.[18,19]

Several explanations have been postulated to specify the role of leptin in the pathogenesis of preeclampsia.[6,7] One hypothesis is that the over-expression of placental leptin and leptin receptors triggers a noradrenaline turnover within the brown adipose tissue so that sympathetic activity is increased in the foetal-maternal unit, stimulating foetal wastage and sudden intrauterine demise.[20,21] Although the exact mechanism is still unclear, animal studies have documented that

prolonged leptin infusion upregulates its receptors in the cardiovascular structures and the central nervous system so that hypertension is initiated.[20,21]

Anato et al conducted a study investigating the effects of antihypertensive treatment on plasma leptin levels of preeclamptic women. The study involved 30 normal pregnant women and 23 pregnant women with severe preeclampsia who had statistically similar values of age, BMI, gestational age and pre-treatment serum leptin concentrations. One hour after the administration of a single dose of alpha-methyldopa or hydralazine alone, or in combination, the serum leptin levels decreased significantly in the treated women.[15] In the present study, pre-treatment plasma leptin levels were significantly higher in the severe preeclampsia group (n = 20) compared to the mild preeclampsia (n = 37) and control (n = 46) groups. The groups were matched in age, BMI and gestational age. Seven days after alpha-methyldopa, or a combined alpha-methyldopa and nifedipine treatment, the plasma leptin levels in the mild and severe preeclampsia groups were reduced to the levels of those in the control group.

To our knowledge, this is the first study held in a population of Turkish women. Although the present study was limited by the relatively short duration of antihypertensive treatment, the significant statistical differences between the study groups favoured a significant reduction of leptin levels using antihypertensive treatments. The present study also yielded data about the foetal status in preeclamptic pregnancies treated with antihypertensive agents. The women with a normal pregnancy, and those with mild and severe preeclampsia were found to be statistically similar in estimated foetal weight, IUGR and foetal demise. This finding may be attributed to the relatively low number of reviewed pregnancies; a larger cohort may have yielded different results favouring antihypertensive treatment.

Alpha-methyldopa and nifedipine are two antihypertensive drugs most commonly administered to treat preeclampsia.[1] Alpha-methyldopa is a centrally-acting sympatholytic agent which may decrease leptin levels by impairing the association between leptin and its receptors within the central nervous system. Nifedipine lowers the BP by blocking calcium channels located on the neurons and cardiac and vascular smooth muscles. Both alpha-methyldopa and nifedipine cross the placenta so that they may interfere with the placental leptin release. It is possible that the leptin released from the placenta stimulates central receptors which regulate the BP and/or the heart rate resulting in hypertension, which in turn causes placental hypoxia and more leptin to be released. Although this pathway resembles a vicious circle, the exact mechanism underlying the increased plasma leptin levels in preeclampsia and the functional role of leptin in the development of hypertension still await further clarification. Subsequent studies should focus on the utilisation of leptin as a marker for screening women who are at high risk for preeclampsia.

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