RELATIONSHIP BETWEEN SERUM LEPTIN AND THYROID HORMONES IN INFANTS WITH TRANSIENT TACHYPNEA OF THE NEWBORN

YENİDOĞANIN GEÇİCİ TAKİPNESİ OLAN BEBEKLERDE SERUM LEPTİN DÜZEYLERİ VETİROİD HORMANLARI İLİŞKİSİ

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ABSTRACT

Purpose: *Leptin is a good example of extreme functional pleiotropy. Originally identified by its effects on food intake and body weight regulation, leptin has subsequently has been shown to play a role in a variety of biological events, including thyroid hormone synthesis and lung maturation. We investigated the serum leptin concentration and its relation to thyroid hormones in respiratory distressed and healthy newborns.* ***Methods:*** *The subjects were 21 infants diagnosed with transient tachypnea of the newborn (TTN) and 28 healthy infants, all delivered by cesarean section. Serum leptin and thyroid hormone levels were measured in the first four hours following delivery. Neonatal gender, birth weight, gestational duration, and antenatal corticosteroid administration were recorded. Intergroup comparisons were made in serum leptin and thyroid hormone levels and the relationship between leptin and thyroid hormone levels was investigated.* ***Results:*** *Serum leptin levels were lower in the TTN group (2.79 ± 0.55 ng/ml) than in the control group (5.53 ± 0.65 ng/ml)(p=0.01). Serum thyroid stimulating hormone (TSH) (p<0.001) and free thyroxine levels (fT4) (p<0.001) were also lower in the TTN group than in the control group and there were positive correlations between serum leptin levels and TSH (r=0.567, p=0.001) and fT4 levels (r=0.484, p=0.001).* ***Conclusion:*** *Hypoleptinemia is associated with TTN in newborns. We suggest that leptin, together with thyroid hormones, may play a role in the hormonal regulation of lung liquid clearance at birth.*

Key Words: *Leptin, Thyroid hormones, Respiratory distress, Newborn.*

ÖZET

Amaç: *Leptin fonksiyonel çeşitliliği olan bir hormondur. İlk olarak yiyecek alımı ve vücut ağırlığının düzenlenmesi üzerindeki etkileri farkedilmiştir. Ancak daha sonra tiroid hormon sentezi ve akciğer maturasyonu da dahil olmak üzere çok çeşitli biyolojik olaylarda rolü olduğu gösterilmiştir. Bu çalışmada, solunum sıkıntısı olan ve sağlıklı yenidoğanlarda serum leptin düzeyleri ve bunun tiroid hormanları ile olan ilişkisi araştırılmıştır.* ***Yöntem:*** *Hepsi sezaryen ile doğan 21 yenidoğanın geçici takipnesi tanısı alan ve 28 sağlıklı bebek çalışmaya alındı. Serum leptin ve tiroid hormon düzeyleri doğumu izleyen ilk 4 saat içinde ölçüldü. Cinsiyet, doğum ağırlığı, gebelik süresi ve antenatal kortikostedroid kullanımı belirlendi. Serum leptin ve tiroid hormonları gruplar arasında karşılaştırıldı, leptin ile tiroid hormonları arasındaki ilişki araştırıldı.* ***Bulgular:*** *Yenidoğanın geçici takipnesi olan grupta serum leptin düzeyleri (2,79±0,55ng/ml) kontrol grubuna göre (5,53±0,65 ng/ml) belirgin olarak düşüktü (p=0,01). Serum tiroid stimulan hormon (TSH)(p<0,001) ve serbest tiroksin (fT4) (0.001) düzeyleri de yenidoğanın geçici takipnesi olan bebeklerde kontrollere göre düşük bulundu. Serumleptin düzeyi ve TSH (r=0,567, p=0,001) ve fT4 (r=0,484, p=0,001) düzeyleri arasında pozitif korelasyon saptandı.* ***Sonuç:*** *Yenidoğanın geçici takipnesi ile hipoleptinemi birarada saptanmıştır. Leptinin tiroid hormonları ile birlikte doğum sırasında akciğer sıvı klirensinin hormonal düzenlenmesinde rolü olabileceği düşünülmüştür.*

Anahtar Kelimeler: *Leptin, Tiroid Hormonları,Respiratuar Distres, Yenidoğan.*

INTRODUCTION

Leptin is a product of the ob gene and provides the brain with information about the fat stores of the body, acting as a feedback signal to inhibit further food intake (1, 2). Shortly after leptin was discovered, it was thought to be the key to understanding obesity. However, it was soon realized that leptin was more than a satiety hormone alone. It exerts an influence in many different physiologic events, including food intake, thermoregulation, adrenal function, sympathetic nerve activation, renal function, blood vessel tone, fertility, immune response, thyroid function and finally lung maturation and central respiratory control (2-8).

Leptin has been shown to play a role in the regulation of thyroid hormone synthesis. Decreased leptin secretion results in suppression of the thyroid axis (9).

Additionally, thyroid stimulating hormone (TSH) has been shown to stimulate leptin secretion by human adipose tissue.

Therefore, it seems that leptin and the thyroid axis maintain a complex and dual relationship (10). Leptin and its receptor are expressed by fetal lung fibroblasts and type II cells and lepin stimulates surfactant synthesis (6, 7). Studies focusing on the role of leptin in lung development have been performed in normal lungs (6, 11). However, no study has evaluated the role of leptin in different pathologic pulmonary conditions. Transient tachypnea of the newborn (TTN) is a common cause of respiratory distress in the newborn. Although delayed resorption of the fetal lung fluid after delivery has been widely regarded as the main problem, the exact cause of inadequate clearance of lung liquid is not known (12). Insufficient b-adrenergic stimulation and thyroid dysfunction are suggested as possible causes of TTN (12, 13).

We measured serum leptin and thyroid hormone concentrations in newborn infants with TTN and compared them with those in healthy newborns. This is the first prospective study investigating serum leptin concentrations and their relation to thyroid hormones in respiratory distressed and normal infants.

MATERIALS AND METHODS

The protocol for the study was approved by the local ethics committee and informed consent was obtained for all subjects. Twenty-one consecutive infants diagnosed with TTN (group 1) and 28 randomly selected healthy newborns (group 2) were included in the study. All had been delivered by cesarean section.

The mean (– SD) gestational age was 36.3 (– 1.7) weeks in group 1 and 36.8 (– 1.8) weeks in group 2. Birth weights varied between 1820 g and 3930 g (mean – SD, 2826 – 553 g) in group 1, and 1550 g and 4360 g (mean – SD, 2811 – 671 g) in the control group. The diagnosis of TTN was reached according to Rawlings (14), depending on: (1) onset of tachypnea (respirations exceeding 60/min) within six hours of birth; (2) persistence of tachypnea for at least 12 hours; (3) chest roentgenogram abnormalities consistent with TTN, such as mild overaeration, vascular congestion, prominent perihilar interstitial markings, and mild cardiomegaly; and (4) absence of other disorders likely to cause similar clinical findings. Infants with documented hypoglycemia, hypocalcemia, polycythemia, meconium aspiration, hyaline membrane disease, sepsis, and congenital pulmonary and cardiac disease with the exception of patent ductus arteriosus were excluded.

Venous blood samples for leptin and thyroid function tests were obtained by venipuncture within 4 hours of delivery, and serum samples were immediately frozen and stored at -30 C until assay after centrifugation. Samples were thawed in a refrigerator prior to analysis. In addition, complete blood count and arterial blood gas values were assessed in all infants. Chest X-rays were obtained from neonates suffering from respiratory distress.

Leptin levels were measured in duplicate using a 125 I Radioimmunoassay (Sensitive Human Leptin RIA Kit, Linco Research Inc, St. Charles, Mo, USA) and samples for quality control were included in each assay. The intraassay coefficient of variation for the assay was 5.9% over the sample concentration range. Thyroid hormones (TSH, T4) were measured by chemiluminescence (DPC Immulite 2000). Blood gas analyses were performed by Nova Plus autoanalyzer (Nova Corpfi, USA).

A chi-square test was used for comparing nominal variables between the groups and a

Mann-Whitney U test was used for the numeric variables. Pearson correlation was used to assess the concordance between leptin levels and other parameters. Multinomial logistic regression analysis was performed to evaluate the effect of gender on leptin between the groups. All statistical calculations were performed using SPSS 10.0 for Windows. A p value of <0.05 was considered statistically significant.

RESULTS

The demographic features of the subjects are shown in table 1. There was no difference between the groups regarding their demographic features. There was a trend for gender distribution between the groups but it was not statistically significant. Male dominance was observed in the TTN group as expected.

The mean (– SEM) serum leptin concentration was 2.79 – 0.55 ng/mL in the infants with TTN, which was lower than that in the healthy infants, 5.53 – 0.65 ng/mL (p=0.01). There were positive correlations between serum leptin levels and gestational age (r=0.347, p=0.01) and birth weight (r=0.337, p=0.02) in all infants. There was no difference between serum leptin levels in girls and boys (p=0.15). Multinomial logistic regression analysis confirmed that the difference in leptin concentrations between the groups was not due to the gender imbalance between the groups.

The mean (– SEM) serum TSH concentration was 16.25 (– 2.34) U/mL and serum fT4 concentration was 1.73 – 0.41 ng/dL in the infants with TTN, which were lower than the concentrations in the healthy infants, 30.54 (– 2.36) U/mL (p<0.001) and 2.63 – 0.20 ng/dL (p<0.001), respectively. There were significant correlations between serum leptin levels and TSH (r=0.567, p=0.001) and free T4 (r=0.484, p=0.001) concentrations in all infants.

The pH value (7.35 – 0.2) and serum total protein concentrations (4.99 –0.79 g/dL) of infants with TTN were significantly lower than those in the healthy babies, with 7.42 – 0.2 for pH and 5.78 – 0.79 g/dL for total protein (p values 0.04 and 0.005, respectively). Other biochemical and hematologic parameters were similar in the two groups.

DISCUSSION

The principal finding of this study was that leptin and thyroid hormones were lower in infants with TTN than in healthy newborns. Furthermore, there was a significant correlation between serum leptin and TSH and free T4 levels, suggesting a potential role of leptin in the regulation of the thyroid axis in newborn infants.

TTN is known as the best described consequence of inadequate neonatal lung liquid clearance (15). Although delayed resorption of the fetal lung fluid during the adaptation to extrauterine life is regarded as the main problem, the exact causes of this maladaptation are generally unknown. Lung liquid clearance is associated with the surge in fetal catecholamines acting via b-adrenoreceptors located in alveolar type II cells and driven by active sodium absorption by increased sodium channel and Na+,K+-ATPase activity (15). Gowen et al. (16) showed that newborn infants with TTN had a transient decrease in amiloride sensitive nasal epithelial Na+ transport compared with normal newborns, supporting the notion that suboptimal clearance of liquid is a cause of TTN.

Table-1: The demographic features of infants in groups 1 and 2

* mean – standard deviation, F/M: female/male, E/G: epidural anaesthesia vs general anaesthesia

	TTN group n=21 (mean – SD)	Control group n=28 (mean – SD)	p value
Birth weight (grams)	2826 – 553	2811 – 671	0.976
Gestational age (wks)	36.3 – 1.7	36.8 – 1.8	0.288
Sex (F/M)	7/14	15/13	0.156
Apgar score (5th min)	9 – 1	9 – 1	1.000
Maternal anesthesia (E/G)	9/11	11/15	0.546
Antenatal corticosteroid administration	4	5	0.600
Cesarean delivery without preceding labor	10	12	0.483

The finding that delivery by cesarean section slowed lung liquid clearance, particularly if undertaken before the onset of labor, indicated that labor was in some way important in the process (17). The association of birth with associated b-adrenergic stimulation and lung liquid clearance was shown by Walters et al., who discovered that epinephrine infusion caused the rapid absorption of lung liquid in mature fetal lambs (18). Labor was shown also to be crucial for the activation of the lung epithelial Na+,K+-ATPase (19). It seems probable that the epinephrine-induced activation of apical Na+ channels and the resulting increase in transepithelial Na+ transport are largely responsible for lung liquid clearance at birth.

The pivotal role of thyroid and glucocorticoid hormones in the maturation of the absorptive response to epinephrine is evident from a series of experiments by Barker and coworkers (20), whose initial studies showed a profound blunting of the response to epinephrine in thyroidectomized fetal lambs that was reversible by infusion of triiodothyronine alone (21). Additionally, regulation of the Na+, K+-ATPase by thyroid hormone has been demonstrated (13). It is suggested that the high degree of coordination of development of the lung liquid clearance and surfactant systems for efficient lung adaptation at birth is regulated by thyroid and steroid hormones synergistically (13, 15).

The present study envisages for the first time the possibility that leptin is involved in the hormonal regulation of lung liquid clearance during postnatal adaptation. However, the mechanisms whereby leptin affects lung liquid clearance are still a matter of speculation. Several possible explanations should be considered. First, leptin may induce epithelial sodium transport by increasing sympathetic outflow. It has been clearly shown that leptin infusion increases sympathetic nerve activity to the various tissues (22, 23).

A second possibility is that leptin may be involved in the regulation of lung liquid clearance by increasing thyroid hormone synthesis. It is known that there is a relationship between leptin and the thyroid gland and it has been suggested that systematically administered leptin in rats stimulated growth and secretion of the thyroid gland through a direct mechanism involving leptin receptors (24, 25). TSH and free T4 levels were also found decreased in infants with TTN and significantly correlated with leptin in the present study, which is consistent with this explanation.

A third possibility is that leptin may act via leptin receptors found in alveolar type II cells, which were recently discovered (7). Bergen et al. (7) have demonstrated that the lung as a whole and fetal type II cells in particular express functional leptin receptors and respond to leptin stimulation by increasing a specific marker for pulmonary surfactant and suggested that leptin may play a role in pulmonary maturation. However, leptin has been shown to inhibit Na+,K+ pump function in 3T3-L1 fibroblasts and suggested to have natriuretic/diuretic effects (26, 27). Thus, it is necessary to investigate whether leptin has any effect on the Na+, K+-ATPase activity in type II cells in the lung.

As a result, one or more of these hypotheses may explain the possible role of leptin in the hormonal regulation of lung liquid clearance at birth. While the results of this study are insufficient for a conclusive implication, they are nevertheless remarkable enough to lead to further studies on the influence of leptin and thyroid hormone interactions on pulmonary transepithelial ion transport at birth.

It has been reported that umbilical venous and arterial leptin levels were higher in babies delivered vaginally than in babies delivered by elective cesarean section, and placental leptin release was suggested to be augmented during advanced labor (28). This is consistent with the risk of TTN being greater after delivery by elective cesarean section. Thus, we selected control subjects from among healthy infants born by cesarean section, and the ratios of the presence of preceding labor were equal in the two groups to eliminate any effect of mode of delivery on serum leptin levels.

There was a trend towards gender distribution between the groups but it was not statistically significant. Male dominance was observed in the TTN group as expected. However, other studies have yielded conflicting results regarding sex differences in terms of leptin in the neonatal period. Some studies have reported that umbilical cord leptin levels were higher in girls than in

boys, whereas other studies have found no gender differences in terms of cord blood leptin in neonates (29-32). We found no sex difference in terms of leptin in infants and concluded that this male dominance had no effect on the results of the present study.

Maternal antenatal steroid treatment increases neonatal plasma leptin (32). In the present study there was no difference between the groups' antenatal corticosteroid exposure and no effect of maternal steroid treatment on neonatal leptin levels was demonstrated.

The pH values of infants were significantly lower in the TTN group than in healthy newborns, but were still within normal ranges. The arterial partial oxygen pressures and Apgar scores of babies in both groups were also normal. Fetal leptin was high in pre-eclampsia and this was attributed to fetal stress such as hypoxia (33). However, Raff et al. (34) reported that hypoxia caused decreased plasma leptin in rats. Thus, as we did not demonstrate profound hypoxemia in any of the infants, we did not suggest that the decreased leptin concentrations in infants with TTN is related to hypoxia.

Although leptin has been shown to play a role in the regulation of thyroid hormone synthesis, the results of clinical studies on leptin and thyroid hormone status are very confusing (9, 24). Moreover, no study has evaluated the relationship between leptin and thyroid hormones in the newborn period. This is the first study suggesting that a possible relationship between leptin and thyroid axis also exists in newborn infants.

In conclusion, we showed that leptin concentrations are decreased in infants with TTN, and hypothesize that leptin, together with thyroid hormones, may play a role in the hormonal regulation of lung liquid clearance at birth.

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