

Lack of Association Between Plasma Leptin Levels and Appetite in Children With Iron Deficiency

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A negative correlation between leptin and appetite or food intake has been shown in healthy individuals. However, the role of leptin in clinical conditions characterized by anorexia has not been established. One of the well-known clinical features of iron-deficiency anemia is poor appetite. We examined the changes in plasma leptin levels in relation to expected improvement in appetite with iron treatment in children with iron deficiency. In 24 infants and small children (mean age \pm standard deviation = 19.6 ± 7.7 months) with iron deficiency, we studied plasma leptin levels before and after iron therapy. After 15.0 ± 2.4 wk of iron treatment, serum ferritin levels improved significantly, with accompanying increases in their subjective appetite scores and food intakes. However, as their mean age and plasma leptin levels adjusted their body mass indexes were unchanged. Serum ferritin correlated significantly with appetite score ($r = 0.680$, $P < 0.001$) and food intake ($r = 0.480$, $P < 0.01$). Leptin correlated only with body mass index ($r = 0.405$, $P < 0.01$). Lack of association between plasma leptin levels and degree of appetite in iron-deficient children treated with iron suggests a leptin-independent mechanism for the observed increase in appetite. *Nutrition* 2001;17:657–659. ©Elsevier Science Inc. 2001

KEY WORDS: leptin, appetite, ferritin, iron deficiency, children

INTRODUCTION

Leptin is thought to act as an afferent satiety signal that regulates weight by suppressing appetite and stimulating energy expenditure in humans.¹ Genetic leptin deficiency produced a dramatically increased appetite and resultant obesity in humans.² Treatment of such an individual with recombinant leptin reduced appetite and caused marked weight loss.³ A negative correlation between leptin and appetite or food intake was shown in healthy adults.^{4–5} In keeping with that concept, plasma leptin should be elevated in clinical conditions characterized by anorexia. However, studies in patients with anorexia nervosa⁶ or inflammatory⁷ or infectious⁸ diseases characterized by inherent anorexia did not show such an inverse relationship. In those studies, profound changes in known confounding factors for circulating leptin levels such as body adiposity⁹ and certain cytokines^{10–11} might have caused this failure.

Poor appetite is a well-known characteristic of iron deficiency (ID). Iron treatment increases appetite in children with ID anemia.¹² This was clearly documented in a study by Lawless et al. who showed that provision of iron supplements improves appetite in terms of recorded food intake and reported level of appetite as compared with children receiving placebo.¹³ We thought that ID would be a better anorectic human model to explore the involvement of leptin in appetite regulation because 1) it is a well-known human condition characterized by poor appetite, 2) restoration of body iron stores is accompanied by enhanced appetite, and 3) it is a relatively benign condition without significant changes in known leptin-influencing factors such as body adiposity, certain cytokines, and toxic metabolites.

We hypothesized that children with ID had increased plasma

leptin levels and improving appetite with iron supplementation was associated with a reciprocal decline in circulating leptin.

MATERIALS AND METHODS

Consecutive infants and small children attending our general pediatric outpatient clinic with the chief complaint of poor appetite were investigated for ID. Definition of ID was based on a serum ferritin concentration below 16 ng/mL.¹⁴ Additional inclusion criteria were a body mass index (BMI) within normal age- and sex-appropriate limits and no acute or chronic illness. Thirty-one infants and small children who met the inclusion criteria constituted the study population. Appetite levels of the subjects were rated by their mothers on a scale of 1 to 10 at the beginning and end of the iron therapy. In addition, the mothers were asked to complete a 3-d food record immediately before and after the study period. Obtained food records were quantified for daily average caloric intake. Plasma leptin levels were measured with an immunoradiometric assay from fasting blood samples obtained between 8 and 12 AM with the use of a commercially available kit (DSL, Webster, TX, USA).

Iron was supplemented as a 20% oral ferrous sulfate solution at an elemental iron dose of $6 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{d}^{-1}$ in three, equally divided doses until the normalization of hemoglobin and ferritin values on monthly follow-ups. Blood samples were obtained 4 mo after the beginning of iron treatment even if the criteria were not met by then. The Wilcoxon signed-ranks test was used to compare the values before and after iron treatment. A general linear-model analysis was used for age and BMI adjustments on plasma leptin. Correlations between the parameters were explored with Pearson's correlation coefficients.

This study was approved by the institutional ethics committee of Mersin University. Informed consents were obtained from the parents of the subjects.

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TABLE I.

MEAN WITH STANDARD DEVIATIONS (RANGES) FOR IMPORTANT STUDY PARAMETERS BEFORE AND AFTER IRON SUPPLEMENTATION IN 24 INFANTS AND SMALL CHILDREN WITH IRON DEFICIENCY

	Before iron therapy	After iron therapy	<i>P</i>
Hemoglobin (g/dL)	9.7 ± 1.5 (5.5-11.7)	11.9 ± 1.1 (9.0-14.1)	<0.001
Serum ferritin (ng/mL)	5.4 ± 3.6 (0.5-12.5)	21.2 ± 11.4 (3.6-48.0)	<0.001
Plasma leptin (ng/mL)	1.5 ± 0.9 (0.3-3.8)	1.8 ± 1.7 (0.2-7.3)	>0.05
Body mass index	16.9 ± 1.5 (14.1-19.2)	17.5 ± 2.1 (15.1-24.8)	>0.05
Appetite score (1-10)	3.3 ± 1.2 (1-5)	6.6 ± 1.3 (4-9)	<0.001
Food intake (kcal · kg ⁻¹ · d ⁻¹)	76.6 ± 12.0 (60-105)	88.1 ± 13.3 (65-120)	<0.001

RESULTS

Seven patients were lost to follow-up during the course of the study. Of the remaining 24 subjects, 14 were boys and 10 were girls, with a mean age of 19.6 (±7.7 standard deviation; range = 8–36) mo. After iron treatment for 15.0 ± 2.4 (range = 8–17) wk, all subjects had improved serum ferritin levels. However, serum ferritin levels were still low in seven subjects at the end of the study. Of note, 20 of the children were diagnosed with ID anemia based on hemoglobin concentrations below the third centile for this age group (11.2 g/dL).¹⁵ Four of them were still anemic at the end of the study despite improvement in their serum ferritin levels. Failure to achieve normal hemoglobin and/or ferritin concentrations was probably due to suboptimal compliance with the iron therapy based on monthly interviews with the mothers. Subjective appetite scores improved in 21 and remained the same in 3 subjects. Daily average caloric intake increased in 21, remained the same in 1, and decreased in 2 children. BMIs were increased in 14, remained the same in 1, and decreased in 9 patients. Plasma leptin levels increased in 11 and decreased in 13 subjects.

Data regarding important study parameters before and after iron supplementation are shown in Table I. The mean age and BMI-adjusted plasma leptin levels before and after iron supplementation did not differ.

Leptin correlated significantly only with BMI ($r = 0.405$, $P < 0.01$). Appetite level correlated significantly with ferritin ($r = 0.680$, $P < 0.001$), hemoglobin ($r = 0.542$, $P < 0.001$), and food intake ($r = 0.515$, $P < 0.001$). Serum ferritin correlated significantly with hemoglobin ($r = 0.562$, $P < 0.001$), appetite score ($r = 0.680$, $P < 0.001$), and food intake ($r = 0.480$, $P < 0.01$).

DISCUSSION

Poor appetite was not associated with increased leptin levels in children with ID and treatment of ID did not cause leptin levels to decline, despite greatly improved appetite and food intake, thus disproving our hypothesis. However, a significant correlation was observed between ferritin, appetite, and food intake. Leptin correlated only with BMI.

The association between appetite and leptin has been clearly demonstrated in humans. Larsson et al. found a negative correlation between leptin and habitual food consumption in 64 healthy adult women.⁴ In another study, Heini et al. found that leptin had a significant negative relation with satiety.¹⁶ Similarly, in a study on the relation between leptin and eating behavior, Keim et al. demonstrated that the greatest desire to eat was associated with the largest percentage drop in plasma leptin levels.⁵

Such an inverse relation was not observed in anorectic conditions as in this study. If leptin was involved in clinical anorexia, those with anorexia nervosa, the most striking anorectic condition in humans, would have increased leptin concentrations after the elimination of the effect of body adiposity. However, patients with

anorexia nervosa have been shown to have low leptin levels.¹⁷ Moreover, Grinspoon et al. explicitly demonstrated that leptin correlated with BMI but not with caloric intake in patients with anorexia nervosa.⁶ In several other anorectic conditions, leptin associations have been studied. Leptin was found to be elevated in patients with chronic renal failure in association with declining glomerular-filtration rates.¹⁸ Those investigators commented that increased leptin might account for poor appetite in uremic patients. However, various factors including urea per se and increased concentrations of many other substances also might be operational in anorexia of uremia.¹⁹ Likewise, increased leptin concentrations induced by certain cytokines such as interleukin-1 and tumor necrosis factor in experimental inflammatory situations have been claimed to be responsible for anorexia of inflammation.¹⁰⁻¹¹ Nevertheless, that idea was not supported by two separate studies carried out in subjects with inflammatory-bowel disease⁷ and acquired immunodeficiency syndrome.⁸ In those studies as in the present one, leptin was associated only with BMI.

We particularly chose our study population to be of normal BMI and free of acute or chronic inflammation because abnormal body adiposity and elevated inflammatory cytokines could blur the picture of the relation between leptin and appetite in this clinical setting. In addition, inflammation would elevate serum ferritin levels, thus disturbing its reliability in representing the labile-tissue iron content.¹²

It has been already shown that ID and its remediation have repercussions in the central nervous system, where appetite is regulated through a partly understood, complex system.¹ For instance, the poor performance of iron-deficient anemic infants on the Bayley scales of mental and motor development was improved to the level of performance of iron-sufficient infants by treatment with ferrous sulfate.²⁰ In another study, iron supplementation improved verbal learning and memory in non-anemic, iron-deficient adolescent girls.²¹ Thus, increased appetite with improved serum ferritin, as shown in this study, might be another reflection of tissue iron changes in the brain.

In conclusion, this study suggests that iron influences appetite regulation independent of leptin and, when taken with the current literature, that leptin is unlikely to be involved as an appetite mediator in ID.

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