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PAPER

Leptin resistance is associated with extreme obesity and aggregates in families

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OBJECTIVE: To examine the determinants of plasma leptin levels and leptin resistance in a sample of extremely obese subjects and their relatives.
DESIGN and METHOD: We obtained plasma leptin values on 968 individuals from 218 families having both extremely obese and average weight members for obesity related variables. Multivariate regression analyses were used to identify predictors of both plasma leptin concentration and an index of leptin resistance. Family correlations and heritabilities were computed for plasma leptin and indices of leptin resistance.
RESULTS: Body mass index and sex predicted 63% of the variance in plasma leptin. Extremely obese subjects were most likely and average weight subjects least likely to be leptin resistant. Both leptin and leptin resistance aggregated within families.
CONCLUSION: Leptin resistance is strongly associated with extreme obesity and appears to be a heritable trait. The determination of its genetic causes will aid in understanding the role of leptin in common forms of human obesity.
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Keywords: body mass index; familial risk; leptin resistance

Introduction

Leptin is a hormone secreted by adipose tissue that acts in the central nervous system as a negative feedback signal to regulate appetite, metabolism and sexual maturation.[1,2] Extremely obese individuals have high leptin levels, which suggests they are resistant to its anorectic and metabolic effects.[1] Meta-analyses support linkage between obesity and the region containing the leptin gene.[3] Further, several studies examining polymorphisms in the 5′ region of the leptin gene and obesity reported positive associations.[4–6] Together, these studies indicate that leptin may play a role in the development of common forms of obesity.

Materials and methods

Subjects

We studied 968 individuals from 218 families ascertained from ongoing genetic studies of obesity at the University of Pennsylvania. A detailed description of the characteristics of the study participants was presented in a previous paper.[7] To be included in the study, probands must have a body mass index (BMI; weight (kg)/height (m^2)) greater than or equal to 40 kg/m^2, have one or more siblings with a current BMI greater than or equal to 30 kg/m^2, one or more siblings must have a maximum lifetime BMI less than 27 kg/m^2, and at least one parent must have a maximum lifetime BMI less than 27 kg/m^2.

Obesity phenotypes

We directly measured weight, height, waist and hip circumferences, and percentage body fat (bioelectrical impedance; Valhalla Scientific). The waist-to-hip ratio (WHR) was computed by dividing the circumference of the waist (cm) by the circumference of the hips (cm). Plasma leptin was assayed using a commercially available radioimmunoassay kit (Linco Research, St Louis, MO).

Leptin resistance

For use as an index of leptin resistance, we computed the ratio of leptin to BMI (LEP/BMI). This index measures leptin level while controlling for the contribution of BMI. We chose BMI rather than percentage body fat or fat mass because in multivariate models, BMI predicted the most variance; also,

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more cases were available for BMI than for percentage fat or fat mass. For qualitative comparisons, a threshold of the 90th percentile of the leptin to BMI ratio (adusted for age within race, sex and generation) was chosen because it yielded an adequate number of cases with a high leptin-to-BMI ratio. In analyses of family correlations, we also examined leptin residualized for BMI.

Statistical methods

We conducted multiple regression analyses to evaluate the relationship between leptin and BMI, percentage fat, fat mass, waist circumference, hip circumference and WHR.

We computed the odds ratios using logistic regression to assess the risk of leptin resistance (LEP/BMI greater than the 90th percentile) using extreme obesity (BMI greater than 40 kg/m^2) and normal weight (BMI < 27 kg/m^2).

We computed familial correlation coefficients for leptin and LEP/BMI, both adjusted for age within race, generation and sex. We also examined the leptin measure residualized for BMI. We applied equal weight to each nuclear family. The correlations were computed using the computer program FCOR2 from the SAGE 4.0 BETA 6 package (http://darwin.c-wru.edu/beta/beta.html). We also estimated heritability of the leptin and leptin resistance measures using the computer program package SOLAR (http://www.sfbr.org/sfbr/public/software/solar/index.html). Adjustment for covariates was completed as described above, ie outside the SOLAR program. In the heritability analyses using SOLAR, we corrected for ascertainment of the primary proband.

Descriptive, linear and logistic regression analyses were computed using the SPSS software (SPSS 9.0 for WINDOWS 2000).

Results

Descriptive and demographic statistics

Mean leptin levels differed by family relationship and by sex within each family relationship (Table 1). As expected, the mean leptin level was highest for probands (59.0 ng/ml) and lowest for fathers (11.9 ng/ml) and brothers (11.7 ng/ml), who also had the lowest BMI. Plasma leptin concentration for females was much higher than for males, in part because most extremely obese individuals were women. No significant difference in leptin levels were observed between European-American and African-American individuals.

Table 1 Mean values of age, body mass index (BMI), and leptin stratified by family relationship and subject characteristics

		Mean value (s.d.)		
Characteristics	*Total*	*Age (y)*	*BMI (kg/m^2)*	*Leptin (ng/ml)*
All subjects	968	47.1 (14.9)	35.6 (10.9)	34.0 (28.9)
Proband	197[a]	39.8 (8.1)	47.6 (8.8)	59.0 (28.8)
Parents	286	65.7 (8.7)	31.7 (8.3)	28.2 (28.3)
Father	105	67.0 (8.5)	29.3 (6.3)	11.9 (12.4)
Mother	181	65.0 (8.8)	33.1 (8.9)	37.6 (30.5)
Sibling	487	39.2 (9.1)	33.1 (9.6)	27.5 (23.5)
Brother	163	39.1 (9.2)	31.1 (8.0)	11.7 (11.8)
Sister	324	39.2 (9.0)	34.1 (10.2)	35.5 (23.8)
Sex				
Male	281	49.6 (16.2)	31.0 (7.9)	12.7 (13.0)
Female	687	46.1 (14.1)	37.5 (11.4)	42.7 (29.1)
Race				
European-American	775	47.9 (15.0)	35.7 (11.2)	34.1 (29.3)
African-American	193	43.9 (14.1)	35.4 (9.8)	33.7 (27.2)

[a]Because leptin was available from some early family members but not all probands, the number of probands is less than the number of families.

Linear regression analysis between leptin and other obesity characteristics

In univariate analyses, BMI, hip circumference, and percentage fat accounted for 55, 53 and 49% of the phenotypic variance, respectively. Sex and waist circumference were also predictors of leptin level, accounting for 23 and 32% of the variance, respectively. Although WHR and age were statistically related to leptin values, they explained little phenotypic variance (8 and 1%, respectively). Race was not a significant predictor of plasma leptin levels, accounting for 0% of the variance.

We then applied multivariate models using variables significant in the univariate analysis. A bivariate model including sex and BMI accounted for the most variance ($r^2 = 0.63$). Other models did not account for significant amounts of additional variance. Thus, the model including BMI and sex was the best predictive model.

Risk for leptin resistance

Extreme obesity (BMI ≥ 40) predicted leptin resistance (LEP/BMI ≥ 90th percentile) with odds ratio of 4.12 (CI = 3.29 – 5.16). Normal weight (BMI < 27) predicted the absence of leptin resistance with an odds ratio of 0.13 (CI = 0.01 – 0.20). Sex did not predict leptin resistance (see Table 2).

Table 2 Risk of leptin resistance (LEP/BMI) for family members

Variable	*Total*	*Affected*	*Odds ratio*	*95% CI*
BMI ≥ 40				
Yes	318	61	4.12	3.29 – 5.16
No	643	35		
BMI ≤ 27				
Yes	266	5	0.13	0.08 – 0.20
No	695	91		
Sex				
Female	683	67	1.07	0.85 – 1.35
Male	278	29		

Family resemblance

Parent–offspring and sibling correlations were highest for leptin residualized for BMI, 0.12 ($P < 0.05$) and 0.18 ($P < 0.05$), respectively. Parent–offspring and sibling correlations were positive but low for plasma leptin, 0.07 and 0.06, respectively, and neither differed from zero. Parent–offspring correlations were similarly low for leptin to BMI ratio, 0.05. However, the sibling correlation for leptin/BMI was higher (0.12) and reached statistical significance ($P < 0.05$). Polygenic analysis which controlled for the ascertainment of the primary proband gave significant heritabilities for plasma leptin (0.18), leptin/BMI (0.15), and leptin residualized for BMI (0.16). All heritabilities were significantly different from zero.

Discussion

We found BMI and sex accounted for 63% of the variance in plasma leptin levels, while race, age and indices of body fat distribution accounted for almost none of the remaining variability. The subjects with highest leptin levels were examined to determine the factors associated with this leptin resistance. Leptin resistance appears to be common in the extremely obese but rare in normal weight subjects.

Plasma leptin concentration aggregated in families. Because plasma leptin concentrations are highly correlated with BMI and BMI is heritable, this observation is expected. However, the families studied here were selected for extreme discordance in body weight,[7] and so the familial aggregation of plasma leptin concentration was surprising. Other studies using families unselected for obesity variables[8–10] found additive genetic effects ranging from 39 to 73% for plasma leptin, but the familial similarity for leptin was largely due to the familial aggregation of BMI. When our families discordant for weight were examined, additive genetic effects accounted for 18% of the variance in plasma leptin and 15–16% of the variance in leptin resistance. Because our families were ascertained for extreme obesity and normal weight, family correlations appear to have been suppressed and the true heritabilities may be higher.

In summary, we found that both plasma leptin level and leptin resistance are heritable traits. Plasma leptin levels and an index of leptin resistance were related to extreme obesity.

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