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Sympathetic-leptin relationship in obesity: effect of weight loss

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Abstract

Obese patients have high plasma leptin concentrations that do not induce the expected responses on weight regulation, suggesting a leptin resistance in obesity. Elevated leptin levels are also thought to be related to a high sympathetic nervous system (SNS) activity. This effect could be preserved, lowered, or even abolished in obesity. We planned to investigate the possible association in a longitudinal study. Ninety-five normotensive healthy women, aged 40.4 ± 11.4 years and body mass index of 33.2 ± 2.3 kg/m^2, were studied. Baseline leptin, fat mass, and heart rate variability were measured and included in a 6-month longitudinal study. Body composition was measured by dual-energy x-ray absorption. Time domain heart rate variability, QT dynamicity, and spectral components on ambulatory electrocardiographs were analyzed. Dietary advice was given by a dietitian to the patient (maximum caloric reduction of 30%), and subjects were randomized in 3 treatment groups: sibutramine 10 mg, sibutramine 20 mg, or placebo. At baseline, low frequencies (LF) and the LF–high frequencies (HF) ratio, mainly related to the SNS, were negatively correlated to leptin concentration ($r = -0.30$, $P = .002$ and $r = -0.36$, $P < .001$) and to the leptin–fat mass ratio ($r = -0.28$, $P = .004$ and $r = -0.33$, $P = .0007$), thus explaining 38% of the LF variance and 33% of the LF/HF variance. Diastolic blood pressure was also negatively correlated to leptin concentrations (−0.20, $P = .04$) and to the leptin–fat mass ratio (−0.22, $P = .022$). In contrast, no consistent correlations between leptin and the time domain components related to vagal activity were observed. At 6 months, after completion of the weight loss program, LF significantly decreased (−7.7% ± 7.9%, $P < .001$), whereas HF was higher than the initial value (+20% ± 5.2%). The leptin–fat mass ratio remained negatively correlated to the LF ($r = -0.34$, $P = .030$) and to LF/HF ($r = -0.35$, $P = .021$) values, explaining 21% of the LF variation. None of the pairwise comparisons between the 2 sibutramine groups and the placebo group were statistically significant for heart rate variability. High leptin concentration is associated with low indexes of cardiac SNS activity and with a lower diastolic blood pressure in normotensive obese women. Our results imply therefore that the relationship between leptin and the autonomic nervous system is disturbed in normotensive obese subjects.

1. Introduction

Obese patients have high plasma leptin concentrations related to the extent of their adipose tissue; however, the elevated leptin level does not induce the expected responses [1] on weight regulation, thus suggesting leptin resistance. Increased leptin and insulin levels are thought to increase sympathetic nervous system (SNS) activity [2] and have been implicated in the hypertension seen in obese subjects [3-6]. In animals, long-term leptin infusions may produce a rise in heart rate and arterial blood pressure [5]. However, in animal models of obesity, the leptin effects on the SNS could be preserved, lowered, or even abolished. Therefore, the effects of leptin on body weight may be separated from those related to the SNS. In the Agouti obese mouse, leptin effects on the cardiovascular system and blood pressure regulation are preserved, despite a well-known leptin resistance in these animals [6,7]. Consequently, leptin resistance may involve food intake regulation and affect, or not, SNS activity. In humans, we suspected that obese subjects who do not develop hypertension could have a resistance to the sympathetic excitatory effects of leptin. As the effect of leptin infusions on the SNS cannot be studied readily, we proposed a study of the relationship between leptin concentrations and cardiovascular sympathetic parameters as assessed by heart rate variability (HRV) in a group of normotensive obese subjects before and after weight reduction.

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2. Subjects, materials, and methods

2.1. Patients and study design

Ninety-five healthy women, aged 40.4 ± 11.4 years (mean ± SD), in whom baseline leptin, fat mass and HRV had been measured were included in a cross-sectional study. All subjects were obese with a mean body mass index (BMI) of 33.2 ± 2.3 kg/m^2. They were recruited by the Nutrition Department of the University Hospital of Nancy, France.

Subjects had to have a stable weight (no more than 3-kg loss during the previous 3 months). Lipid-lowering agents (monotherapy only) were only permitted if they had been taken in stable doses for at least 3 months.

Subjects were excluded from the study on the basis of any of the following: serious eating disorders, obesity of endocrine origin, diabetes mellitus, supine diastolic blood pressure >95 mm Hg or supine heart rate >100 beats per minute, any significant past illness including hypertension, electrocardiographic (ECG) or laboratory abnormalities, any cardiac dysfunction, taking of any drug or dietary supplement that might alter body weight or insulin sensitivity, or taking any antihypertensive drug.

The study protocol was approved by the Ethics Committee of Nancy and was conducted in accordance with Declaration of Helsinki and European Good Clinical Practice guidelines.

2.2. Study investigations

Physical examination, a 12-lead ECG, and laboratory assessments were performed at the Clinical Investigation Center. Blood pressure was measured to the nearest 2 mm Hg after 5 minutes of supine posture with a standard sphygmomanometer. Body weight to the nearest 0.1 kg and height (without shoes) were measured with the subject in light clothing. The waist-hip ratio was calculated from measurement of the waist and the hip circumferences, the latter being the largest measured circumference around the buttocks.

Body composition was measured at baseline and at 6 months. Eligible patients were randomized to 3 treatment groups for a period of 6 months (sibutramine 10 mg, sibutramine 20 mg, or placebo prescribed once daily in the morning). For reasons of tolerability, patients in the sibutramine 20-mg group received 15 mg for the first 2 weeks of the double-blind phase, after which the dose was increased to 20 mg. At baseline, dietary advice was given by a dietitian to the patient, which included a maximum caloric reduction of 30%. The sibutramine and matched placebo capsules were provided by Knoll Pharmaceuticals (Abbott, Nottingham, United Kingdom) as part of their partial financial support of this study, but this company was not involved in data analysis or the preparation of this report.

Leptin concentrations were determined at 6 months, and ambulatory ECG monitoring was performed at 6 months.

2.3. Body composition analysis by dual-energy x-ray absorptiometry

Body composition was estimated from a total body scan using a LUNAR DPX-IQ system (LUNAR, Madison, WI) with the 4.1d software version.

2.4. Laboratory analysis

Serum samples were stored at −20°C until assayed. All samples were analyzed at the same time. Insulin resistance was derived using the homeostatic model assessment calculation according to Matthews et al [8], this being fasting insulin (in microunits per milliliter) × fasting glucose (in millimoles per liter)/22.5. Glucose was measured by the glucose oxidase method (Beckman Instruments, Fullerton, CA).

Plasma insulin was determined by immunoassay (Insulin IMX; Abbott Laboratories, Tokyo, Japan) at baseline and at 6 months. The intraassay coefficient of variation was 4%, and the interassay coefficient was 6%. Cross-reactivity with proinsulin was 0.05%.

High-density lipoprotein cholesterol was measured after precipitation of apolipoprotein B–containing lipoproteins with sodium phosphotungstate/magnesium chloride (Boehringer Mannheim, Mannheim, Germany). Total cholesterol and triglycerides were measured using commercial kits (Boehringer) adapted to a Hitachi 911 analyzer (Boehringer Mannheim). Low-density lipoprotein cholesterol was calculated according to Friedewald et al [9]. Serum leptin levels were measured using a radioimmunoassay (Linco Research, St Louis, MO), the intra- and interassay coefficients of variation of which were 4.5% and 8%, respectively.

2.5. ECG and HRV

Ambulatory ECG monitoring at baseline and 6 months was performed using 2-channel frequency modulatory tape recorders. After careful preparation of the skin, 6 electrodes were placed on the chest to obtain the bipolar chest leads. Two independent and blinded experienced investigators analyzed the ambulatory ECG recording tapes by ELATEC (VFC module; 3.02 VF ELATEC logitiel, ELA Medical, Mountrouge, France). Ectopic beats were corrected for linear interpolation with the adjacent complexes. Electrocardiographic tracings with more than 1% premature beats were eliminated from the analysis.

Spectral components were identified and estimated using the spectral decomposition algorithm and were then assigned, on the basis of their central frequency, to 1 of 3 bands: very low-frequency (VLF) band (from 0-0.040 Hz), low-frequency (LF) band (from 0.040-0.150 Hz), and high-frequency (HF) band (from 0.150-0.400 Hz). The power of LF and HF components was expressed in milliseconds squared (absolute units) and, as recommended by the Task Force of the European Society of Cardiology and the North American Society of Pacing and Electrophysiology [10], in normalized units (nu) by dividing the power of each component by the total power less the sum of the ultralow

frequency and the VLF components (0-0.03 Hz) and multiplying by 100. The LH/HF ratio was also computed. Whereas the HF component is considered a pure index of efferent vagal activity, the LF component is influenced by both sympathetic and vagal activity.

For each recording, 24-hour average values of the normal time interval between consecutive heart beats (RR), SD of all normal RR intervals in the entire 24-hour ECG (SDNN), mean of the SDs of all normal RR intervals for all 5-minute segments (SDNNindex), SD of the 5-minute mean values of normal RR intervals (SDANN), square root of the mean of the sum of the squares of differences between adjacent RR intervals (RMSSD), and the number of adjacent normal RR intervals differing by more than 50 milliseconds, as percentage of the total number of normal RR intervals (pNN50), were computed [10].

The QT dynamicity was evaluated by analyzing 24-hour Holter recordings. The linear regression slope of the QT interval measured to the apex (rQTa) and to the end of the T wave (rQTe) plotted against RR intervals was calculated using a dedicated Holter algorithm.

2.6. Statistical analysis

Values are expressed as means ± SD. The BMDP statistical software (University of California Press, Berkeley, CA, 1992) was used to perform the statistical analysis. The normal distribution of frequency was verified by skewness and kurtosis tests. Linear regression analysis was used to evaluate correlations between variables, whereas multiple regression analysis was used to assess the effects of several independent variables on dependent variables.

Weight loss effects were calculated as changes from baseline (M0) to month 6 (M6) and assessed by paired *t* test in each group. Analysis of variance was used to analyze differences among the means of parameter variations between M0 and M6 of the 3 groups. Statistical significance was defined as $P < .05$.

3. Results

3.1. Baseline HRV components and blood pressure

The clinical and biochemical characteristics of the study patients are presented in Table 1. Seventy-nine women had a BMI between 30 and 35 kg/m^2; and 16 women, between 35 and 40 kg/m^2. Fat mass measured by dual-energy x-ray absorptiometry ranged from 26.2 to 58.4 kg; leptin, from 15 to 81.5 ng/mL; and leptin–fat mass ratio, from 0.44 to 1.74 ng/(mL kg). The main indexes of HRV are detailed in Table 2.

3.1.1. Regression analysis

As expected from literature reports, leptin concentration positively correlated with the BMI ($r = 0.29$, $P < .001$), body fat mass ($r = 0.45$, $P < .0001$), waist-hip ratio ($r = 0.32$, $P < .001$), and insulin concentration ($r = 0.25$, $P = .011$). The leptin–fat mass ratio was positively correlated to the percentage of body fat mass ($r = 0.22$, $P < .05$). As shown in Table 3 and Fig. 1, plasma leptin levels and leptin–fat mass were negatively correlated with LF (nu) and the LF/HF ratio (parameters indicative of the sympathetic component and sympathetic/vagal balance, respectively), SDNN (which provides an estimation of overall HRV), and the rQT. The HRV parameters were negatively correlated with age ($P < .05$), whereas the remaining parameters were not correlated with age. In contrast, no consistent correlations between leptin and the time domain components mainly related to the vagal activity (RMSSD, pNN50) were observed (data not shown). Diastolic blood pressure was negatively correlated with leptin concentrations and with the leptin–fat mass ratio (Table 3).

The LF (nu) and LF/HF correlated negatively with insulin concentration ($r = -0.25$, $P = .028$ and $r = -0.19$, $P = .08$, respectively). The LF (nu) was significantly correlated with the leptin–fat mass ratio, age, and insulin, whereas correlations with waist-hip ratio and percentage of body fat were not significant. These LF-leptin relationships remained significant after adjustment for confounding factors (insulin, waist-hip ratio, percentage of body fat, and age) (leptin: $r = -0.24$, $P = .027$; leptin–fat mass ratio: $r = -0.21$, $P = .036$). Heart rate and systolic blood pressure did not show any independent association with leptin or leptin–fat mass ratios.

Multiple linear regressions (Table 4) were performed to assess the independent contribution of age, leptin or leptin–fat mass ratio, percentage of body fat or fat mass, waist-hip ratio, and insulin to the variation in HRV parameters. Partial correlation coefficients for LF proportion, LF/HF, or rQTe, adjusting for the other independent variables, are presented along with r^2 values for the models. The LF proportion was negatively correlated to the leptin–fat mass ratio, insulin concentration, and age. The LF/HF and rQTe were also negatively correlated with the leptin–fat mass ratio. The

Table 1
Clinical characteristics, body composition and distribution, and biological parameters (N = 95)

Variables	Means ± SD	Range
Age	40.4 ± 11.4	19.0-62.0
BMI (kg/m^2)	33.2 ± 2.3	30.0-39.8
Waist-hip	1.17 ± 0.07	1.00-1.32
Leptin (mg/L)	37.3 ± 11.9	15.0-81.5
Leptin-fat (mg/[L kg])	0.90 ± 0.26	0.44-1.74
Insulin (mU/L)	22.5 ± 22.8	3.7-140.0
HOMA	1.03 ± 1.37	0.16-8.88
Blood glucose (mg/dL)	91 ± 14	45-125
Cholesterol (mg/dL)	208 ± 37	120-340
HDL cholesterol (mg/dL)	55 ± 12	29-93
Triglycerides (mg/dL)	136 ± 84	31-402
% fat mass	46.6 ± 3.7	37.5-56.4
Fat mass (kg)	41.5 ± 5.9	26.2-58.4

HOMA indicates homeostatic model assessment; HDL, high-density lipoprotein.

Table 2
Blood pressure, heart rate, HRV, and QT (N = 95)

Variables	Mean ± SD	Range
SBP (mm Hg)	122.8 ± 12.5	89.0-163.0
DBP (mm Hg)	74.4 ± 9.1	55.0-95.0
PUL (beats/min)	72.2 ± 7.9	55.0-93.0
Total power (ms^2)	3258 ± 178	550-8294
LF/HF	5.40 ± 2.39	0.92-12.49
LF power	927 ± 541	98-3150
LF proportion	0.82 ± 0.08	0.48-0.93
RMSSD	33.6 ± 15.4	10.8-80.1
SDANN	123.7 ± 33.1	60.4-228.4
SDRR	116 ± 37	57-209
rQTa	0.92 ± 0.04	0.82-0.98
rQTe	0.84 ± 0.11	0.44-0.96

SBP indicates systolic blood pressure; DBP, diastolic blood pressure; PUL, pulsation; SDRR, SD of all normal RR intervals.

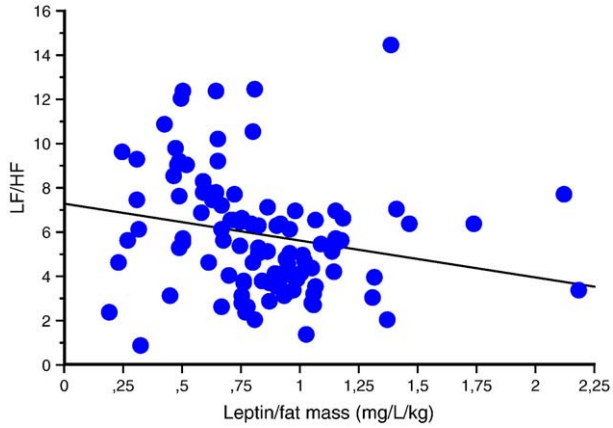

Fig. 1. Regression analysis between leptin–fat mass ratio and LF/HF ratio ($r = -0.21$, $P = .027$).

results shown in Table 4 explain 38% and 33% of the variation of LF and LF/HF, respectively.

The results of these correlations were not significantly different if LF and HF were expressed in milliseconds squared (data not shown).

3.2. Six months' HRV components and blood pressure: changes in body composition and leptin

Fig. 2 shows the mean changes in weight, fat mass, leptin, and leptin–fat mass ratio from baseline to month 6 for each treatment group. At month 6, as expected, weight loss differed significantly among the 3 groups. The subjects who received 20 mg sibutramine lost 10.5% ± 7.2% of their initial weight compared with 7.0% ± 4.4% for the 10-mg sibutramine group or 2.9% ± 3.5% for the placebo group ($P < .001$).

Despite the fact that patients who received 20 mg sibutramine lost more weight than patients receiving the lesser dose, leptin concentrations decreased significantly in both sibutramine groups at month 6, without significant differences being found between the 2 groups.

3.2.1. Effects of weight loss on heart rate and HRV

Changes in total HRV spectral power and other parameters from baseline to month 6 are presented in Table 5. Compared with placebo, patients treated with sibutramine 10 mg had a slightly greater fall in both systolic and diastolic blood pressure from baseline to end point or month 6, although the changes were not statistically significant. For patients in the higher sibutramine dosage group, blood pressure levels increased relative to placebo; but these changes did not reach statistical significance.

At 6 months, total power and HF (nu) significantly increased compared with the baseline values. The LF significantly decreased, whereas HF was about 20% higher than the initial value. Changes in the LF/HF ratio were the opposite, with a significant decrease observed at month 6 compared with the baseline values.

The changes in the LF (nu), HF (nu), and LF/HF ratio from baseline to 6 months were not statistically different among the 3 subgroups treated. None of the pairwise comparisons between the 2 sibutramine groups and the placebo group were statistically significant for these parameters.

Table 3
Correlation between fasting plasma leptin and variables studied at the inclusion

	Leptin		Leptin-FM ratio	
	R	*P*	*r*	*P*
Dependent variables				
SBP	–	NS	–	NS
DBP	−0.20	.040	−0.22	.022
HR	–	NS	–	NS
Frequency domain methods (age adjusted)				
Total power	–	NS	–	NS
LF proportion	−0.30	.002	−0.28	.004
LF (ms^2)	−0.21	.016	−0.25	.027
LF/HF	−0.36	<.001	0.33	.0007
SDRR	−0.33	.026	−0.26	.07
mRR	−0.29	.05	−0.22	NS
rQTe	−0.31	.037	−0.31	.038

FM indicates fat mass; NS, not significant; mRR, mean RR intervals.

Table 4
Multivariate analysis using HRV components and QT as dependent variables at the time of inclusion

		Baseline (N = 95)			
Dependent	Independent	β	F	*P*	r^2
LF (nu)	Leptin-FM	$-50.5 \cdot 10^{-3} \pm 25.1 \cdot 10^{-3}$	4.1	.012	0.38
	Age	$-2.1 \cdot 10^{-3} \pm 0.6 \cdot 10^{-3}$	8.6	.001	
	Insulin	$-1.1 \cdot 10^{-3} \pm 0.35 \cdot 10^{-3}$	10.4	.028	
	% Body fat mass	$-3.4 \cdot 10^{-3} \pm 1.95 \cdot 10^{-3}$			
LF/HF	Leptin-FM	−2.63 ± 0.90	8.5	<.001	0.33
	Age	−0.065 ± 0.238	7.6	<.001	
	Insulin	–			
	% Body fat mass	–			

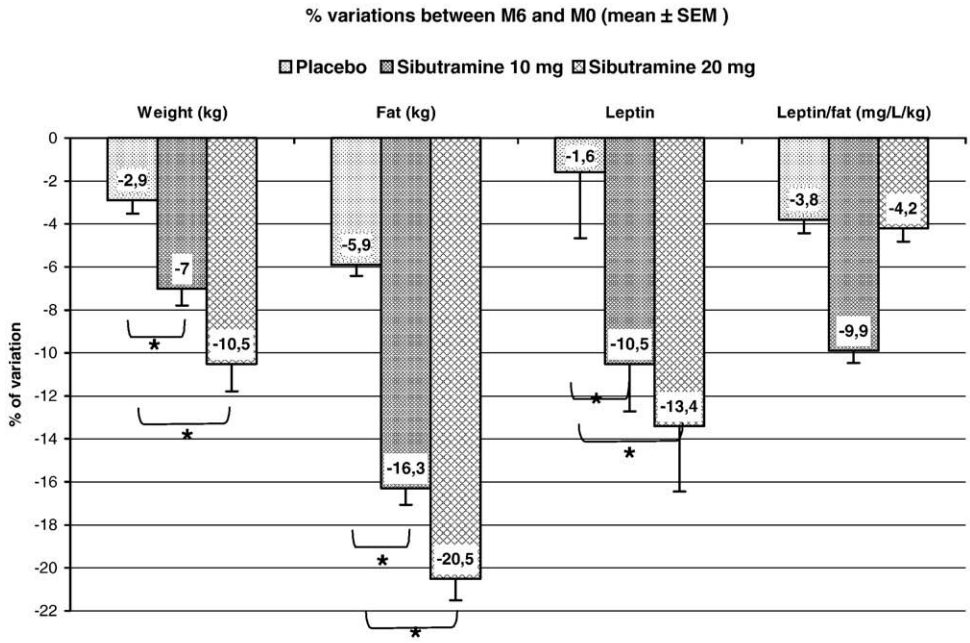

Fig. 2. Evolution of weight, fat mass, leptin concentrations, and leptin–fat mass ratio between M6 and M0 (mean ± SEM) in the 3 treatment groups.

The LF values at month 3 were predictive of the weight loss and fat mass loss (percentage and kilogram, respectively) between the third and sixth months irrespective of the treatment. In fact, fat loss at month 6 was positively and significantly correlated to the LF values at month 3 ($r = -0.27$, $P = .041$ for fat mass loss [percentage]; $r = -0.29$, $P = .034$ for fat mass loss [kilogram]; and $r = -0.25$, $P = .05$ for weight loss [kilogram]). As a result, subjects with low LF at month 3 lost less weight between the third and sixth months than subjects with high LF. However, there was no correlation between Δ weight and Δ LF.

3.3. Relationship between HRV variables and leptin at month 6

Analyses were also performed to assess the contribution of leptin–fat mass ratio, waist-hip ratio, percentage of body fat mass, age, and treatment to the variation of LF (nu) or LF/HF at 6 months (Table 6). Leptin–fat mass ratio and age were inversely associated with LF, explaining 21% of the variation in this variable. A similar result was observed for LF/HF. Sibutramine treatment had no influence on this result. Thus, women who had high leptin levels at month 6 also had a low LF or LF/HF values.

4. Discussion

The novel and important finding in this study is that high leptin is associated with low indexes of cardiac SNS activity and with a lower diastolic blood pressure in normotensive obese women. The LF spectral component, mainly related to sympathetic modulation of the heart rate, and the LF/HF ratio, an index of the sympathovagal balance [11], were both negatively correlated to leptin concentration, independently of body fat mass. A decrease in the time domain measures of HRV, SDNN estimation of overall HRV, and the rQT was also negatively correlated to the leptin level. In contrast, the indexes recognized as being influenced by vagal tone were not associated with leptin concentrations. Interestingly, despite a decrease in both LF (nu) and LF/HF ratio after weight loss, the negative correlation between these indexes and leptin–fat mass ratio remained significant after 6 months of food restriction and/or sibutramine treatment.

These results are opposite to those reported by Paolisso et al [12] in nonobese subjects, where leptin concentration was positively correlated to the LF/HF ratio. Our results imply therefore that the relationship between leptin and autonomic nervous system is disturbed in normotensive obese subjects. These results suggest that such patients could develop a resistance to the sympathoexcitatory effects of both leptin and insulin. However, the reasons for these discrepancies are not clearly understood.

Weight loss did not modify the negative relationship between leptin and sympathetic modulation that was found at baseline. This finding could imply that normotensive obese subjects with high leptin–fat mass ratios were still resistant to the action of leptin on the SNS, even after weight loss.

Despite an increase in the HRV, the relative contribution of the LF (LF [nu]) decreased weakly but significantly at 6 months and was lower than at baseline. In contrast, HF (nu) increased. As previously described, sympathetic nerve activity decreases with weight loss [13]. Of interest, Verwaerde et al [14] reported contrasting results in obese dogs during weight gain. In this animal model, body weight

Table 5
Comparison of heart rate, blood pressure, and parameters of HRV between baseline and the sixth month

	Inclusion	6th month	Means of differences between M6 and M0 (%)	Comparison of the means of differences among the 3 groups (*P* ANOVA)
Heart rate				
Placebo	73.5 ± 9.7	66.7 ± 9.1	−10.3% ± 11.3%***	<.001
Sibutramine 10 mg	71.3 ± 8.3	73.2 ± 8.8	6.9% ± 13.6%*	
Sibutramine 20 mg	73.9 ± 7.6	73.6 ± 9.5	2.2% ± 13.6%	
SBP				
Placebo	120 ± 12	120 ± 9	0.2% ± 9.2%	NS
Sibutramine 10 mg	121 ± 12	118 ± 11	−1.5% ± 9.5%	
Sibutramine 20 mg	122 ± 11	125 ± 13	2.5% ± 8.4%	
DBP				
Placebo	75 ± 8	74 ± 9	−0.4% ± 10.5%	NS
Sibutramine 10 mg	75 ± 9	72 ± 9	−1.5% ± 9.9%	
Sibutramine 20 mg	74 ± 8	76 ± 7	2.8% ± 10.9%	
Total power				
Placebo	3233 ± 2021	4777 ± 3348	62.8% ± 9.0%***	NS
Sibutramine 10 mg	2371 ± 1503	3445 ± 2224	46.6% ± 22.3%***	
Sibutramine 20 mg	3002 ± 1426	4352 ± 2480	42.0% ± 25.7%***	
% LF				
Placebo	76.2 ± 9.4	70.4 ± 13.0	−7.5% ± 7.6%***	NS
Sibutramine 10 mg	72.8 ± 9.3	67.6 ± 13.2	−8.3% ± 10.8%***	
Sibutramine 20 mg	75.7 ± 8.9	70.5 ± 11.1	−7.3% ± 5.4%***	
% HF				
Placebo	23.8 ± 9.4	29.5 ± 13.1	20.1% ± 5.1%***	NS
Sibutramine 10 mg	27.1 ± 9.1	32.4 ± 13.2	20.4% ± 7.3%***	
Sibutramine 20 mg	24.3 ± 8.9	29.5 ± 11.2	19.6% ± 3.1%***	
LF/HF				
Placebo	5.4 ± 1.9	3.2 ± 2.1	−41.5% ± 24.1%***	NS
Sibutramine 10 mg	5.7 ± 2.2	2.8 ± 2.4	−42.2% ± 71.7%***	
Sibutramine 20 mg	5.6 ± 2.1	2.8 ± 1.2	−46.9% ± 24.9%***	

ANOVA indicates analysis of variance.
Significant difference between M0 and M6: *$P<.05$; **$P<.01$; ***$P<.001$.

increment was associated with a transient increase in the LF band. As a result, these changes may indicate a shift of sympathetic-vagal balance toward reduced sympathetic tone and vagal predominance after weight loss. Similar conclusions were obtained by considering changes in the LF/HF ratio. There is growing consensus that these changes can be beneficial and provide cardiovascular protection. In contrast, a decreased HRV was associated with an increased mortality [15]. These results agree with those of Grassi et al [13], who reported improved baroreflex sensitivity in normotensive obese subjects after short-term (6-week) energy restriction.

Patients treated with 10 mg sibutramine had a slightly higher heart rate at month 6 than at baseline. In contrast, the LF component of HRV was markedly reduced with sibutramine as in the placebo group. These paradoxical changes with sibutramine are consistent with previous studies on healthy subjects and suggest a combination of peripheral and central nervous system mechanisms [16,17]. Low-frequency heart rate oscillations, the so-called Mayer waves, are mediated by the SNS. The decrease in the LF component of HRV at 6 months is probably due to the effect of weight loss on the SNS tone because the fall is not significantly different between the placebo and sibutramine groups. Sibutramine did not modify this expected reduction.

One possible explanation for the increased heart rate is that the cardiovascular system becomes more responsive to stimulation of adrenergic receptors by endogenous catecholamines after weight loss.

Relating leptin levels to body fat may help predict leptin resistance [18]. High plasma leptin adjusted for body fat might be considered as a marker of leptin resistance in animal models of obesity as well as in obese humans. This possibility has to be taken into account when analyzing the relationship between leptin and SNS activity. In humans, plasma leptin correlates significantly with blood pressure and muscle sympathetic nerve activity [7]. As a result, leptin may contribute to hypertension in obese patients [19-21], but only if the action of leptin is not attenuated. One other explanation is that leptin resistance could be selective in obese subjects, affecting the weight-reducing effect but not the SNS stimulation. Moreover, studies on an animal model of leptin resistance, the Agouti yellow obese mouse, support this hypothesis. In this model, mice are resistant to the appetite- and weight-reducing effect of leptin; nevertheless, short-term intracerebroventricular administration of leptin increases sympathetic nerve activity to the kidney in a dose-dependent manner without affecting body weight and food intake [6,22]. This contributes to the regulation of blood pressure in these mice [23].

Table 6
Multivariate analysis using HRV components and QT as dependent variables at month 6 of follow-up

6th month (N = 95)					
Dependent	Independent	β	F	*P*	r^2
LF (nu)	Leptin-FM	−0.132 ± 0.057	5.4	.041	0.21
	Age	$-4.1\cdot10^{-3} \pm 1.4\cdot10^{-3}$	7.9	.036	
	% Body fat	–			
	Mass	–			
LF/HF	Leptin-FM	−2.52 ± 0.97	6.7	.049	0.22
	Age	−0.099 ± 0.032	9.7	.011	
	% Body fat	–			
	Mass	–			

Patients with relative hyperleptinemia without hypertension, such as those selected in our study, might represent a group of patients with a nonselective leptin resistance. The negative relationship reported in this study between baseline diastolic blood pressure and leptin–fat mass ratio supports this hypothesis. On the contrary, this relationship was found to be positive in hypertensive obese subjects [20]. It would be very interesting to compare the HRV components of hypertensive and normotensive subjects. It is also known that the association between high sympathetic activity and obesity does not exist in some ethnic populations in which the prevalence of obesity is high and that of hypertension is weak, such as Pima Indians. Spraul et al [24] have demonstrated that sympathetic activity measured by microneurography was significantly related to percentage of body fat in white persons, but not in Pimas. Our data are consistent with an alteration of the SNS regulation of both HR and BP at least in some obese normotensive subjects [25].

The rat F-344xBN animal model illustrates this phenotype of obesity [26]. Similar to humans, these aged rats become obese in early senescence despite high serum leptin concentrations. In this model, leptin-induced transcription factor binding diminished with age. After both peripheral and central leptin administration, the decrease in food intake and the increase in energy expenditure were attenuated in the older obese rats in accordance with a reduced responsiveness to peripheral and central leptin in this model.

Leptin effects on vascular tone and blood pressure remain poorly defined in humans. Haynes et al [27] demonstrated that short-term intravenous infusion of leptin increased the level of sympathetic nerve activity to the kidney, adrenal gland, and brown adipose tissue. However, sodium excretion by animals infused with leptin was significantly increased [28]. Hyperleptinemia also rapidly enhanced endothelial function [29]. As a result, the diuretic effect of leptin could relax smooth muscle tone in the peripheral arteries and decrease diastolic blood pressure. These effects, distinct from the sympathetic stimulating effects of leptin, could explain the negative correlation between leptin and diastolic blood pressure in normotensive subjects.

In healthy humans, changes to leptin levels within the physiological range during short-term fasting do not seem to influence LF power or LF/HF balance [16]. Nevertheless, a 72-hour fasting leads to an increase in SNS activity because of the decrease in glycemia, whereas serum leptin levels dramatically decreased. Under these conditions, sympathetic tone is probably high despite low leptin and insulin levels. Secondly, in that study, leptin levels were low even after leptin administration (baseline endogenous leptin concentrations were 37.3 ± 11.9 mg/L in our study vs 29.6 ± 2.2 mg/L after leptin administration in the cited study). As a result, the study by Chan et al [16] does not rule out a leptin stimulating effect on the SNS.

Matsumoto et al [30] showed in a transversal study on subjects that both the ratios of the VLF component (reflecting thermoregulatory sympathetic function) and the global SNS index ([very low + low]/high frequencies) to plasma leptin concentration were markedly reduced in the obese compared with the control group. The study of weight loss effects is a first approach to analyze this relationship in a longitudinal study. The hypothesis that normotensive obese subjects have a global leptin resistance (central and peripheral leptin resistance), whereas hypertensive obese subjects have selective resistance (central), could be studied by a comparison of SNS activity before and after leptin administration in hypertensive or normotensive obese subjects. The assessment of sympathetic activity in humans can be approached using a number of hemodynamic, pharmacological, biochemical, and neurophysiological techniques that may lead to differing conclusions whether a global or tissue-specific index of sympathetic activity is used. A direct measurement of sympathetic activity at the level of the nerve, such as may be obtained using microneurography, could be interesting. However, longitudinal studies are needed to examine the effects of leptin on sympathetic activity and on the development of hypertension.

In conclusion, an assessment of HRV indexes has shown that normotensive obese subjects with high leptin levels have a lower SNS activity than normotensive obese subjects with low leptin levels. Using the leptin–fat mass ratio or statistical adjustment for body fat mass did not eliminate the negative relationship between leptin and HRV components or diastolic blood pressure. Diet-induced weight loss with or without sibutramine dramatically decreased sympathetic indexes of HRV. However, the relationship between leptin and SNS indexes remained inversed at the end of the 6-month weight loss program, suggesting that reduced sympathetic responsiveness to endogenous leptin production, implying peripheral leptin resistance, might be a pathophysiological feature of obesity.

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