VLCD-Induced Weight Loss Improves Heart Rate Variability in Moderately Obese Japanese

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To evaluate the effects of weight reduction on the autonomic nervous system in obese patients, we investigated heart rate variability (HRV) based on 24-hr ambulatory electrocardiogram (ECG) recordings before and after weight reduction. To aim for weight reduction, 16 obese patients were treated with the very-low-calorie conventional Japanese diet (VLCD-CJ) therapy combined with behavior therapy. Percent weight reduction was 17.8% ± 1.5% (means ± SEM), but mean blood pressure did not change significantly after VLCD-CJ therapy. The mean normal R-R interval (mNN) of the 24-hr ECG and all other five time-domain indices increased after weight reduction. Spectral analysis revealed that weight reduction increased the high frequency (HF) component, but decreased the ratio of low to high (LF/HF) components. Rate of change in mNN or HF correlated positively with reduction rate of body mass index, but not that in LF/HF. Analysis of daily fluctuations in each HRV parameter showed that significant improvement after weight loss occurred mainly during the nocturnal period, but an HF component was improved throughout the day and night periods. These findings indicate that functional impairment of the autonomic nervous system in obese subjects, particularly in the nocturnal period, is improved by effective weight reduction after VLCD-CJ therapy. [Exp Biol Med Vol. 226(5):440–445, 2001]

Key words: heart rate variability; obesity; weight reduction; conventional Japanese VLCD therapy; spectral analysis; parasympathetic dominance

Together with hyperinsulinemia and hypertension (1), dysfunction of the autonomic nervous system has been reported to play a role in obesity development (2). Indeed, inactivation of the sympathetic nerves has been found to reduce thermogenesis in various peripheral tissues and adipocyte lipolytic activity, which in turn reduce energy expenditure and accelerate development of obesity (3). An animal obesity model with bilateral lesions of the ventromedial hypothalamic nucleus is well known to induce hyperinsulinemia by increasing vagal stimulation of the pancreatic islets while concomitantly suppressing sympathetic activity (4). In contrast, it has been reported that hypertension and hyper insulinemia in obese humans result from sympathetic rather than parasympathetic activity (1). Such a conclusion, however, may derive from the fact that parasympathetic activity is methodologically difficult to measure in human obesity.

There is a growing body of evidence that analysis of heart rate variability (HRV) is useful for predicting sudden cardiac death in either postinfarction or diabetic patients (5, 6). Fluctuation of heart rate around mean heart rate provides essential and valuable information on impairment of sympatho-vagal activity in the cardiorespiratory regulatory systems (7, 8). One report showed that a 10% body weight gain significantly decreased HRV, which was attributable to decreased parasympathetic activity, whereas a 10% weight loss had no effect (9). However, a blocker study of sequential inhibition of cardiac autonomic nerves demonstrated that a 10% body weight gain reduced parasympathetic activity in non-obese subjects, while a 10% weight loss increased parasympathetic activity and decreased sympathetic activity in both non-obese and obese subjects (10). Such studies provide evidence that pathological autonomic nerve function occurs in obese humans, while differences in experimental design, incomplete methodological assessment, and insufficient data regarding HRV may explain inconsistent results (9, 10).

As mentioned previously, one of the major difficulties in evaluating HRV is the methodological difficulty in assessing sympathetic activity. Nevertheless, HRV is still a beneficial tool for assessment of autonomic nerve function because it enables us to evaluate autonomic nerve function continuously for a 24-hr period, and to assess the function expressed as the balance of autonomic nerve activity. In addition, the measurement procedures *per se* are not harmful to the subjects. In obese animal models, it has been

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demonstrated that diurnal fluctuation in autonomic nerve activity is abnormally desynchronized (11). Disrupted circadian rhythm of autonomic activity contributes to the advance shift of circadian ingestive and ambulatory rhythms (12). The present study was designed to reevaluate the relative advantage of HRV to determine whether more than 10% excessive weight loss in obese subjects may affect daily fluctuations of cardioautonomic parameters, based on 24-hr ambulatory electrocardiography (ECG), and how and what parameters of HRV may adequately reflect changes in autonomic nerve activity following weight reduction.

Materials and Methods

Subjects. Between April 1992 and March 1993, 16 obese Japanese inpatients, including 14 women and two men, between 20 and 60 years of age (mean, 47 ± 3 years old), each with over 25.0 kg/m^2 body mass index (BMI) (mean, 31.4 ± 1.1 kg/m^2), were admitted in Yufuin Kouseinenkin Hospital for further evaluation of obesity complications and were placed on therapy for obesity. They were selected from 36 obese inpatients by excluding those with diabetes mellitus, cardiovascular disorders, or hypertension, as well as those who were smoking or were medicated with drugs affecting autonomic nerve function. A seventy-five-gram oral glucose tolerance test (OGTT) proved that no diabetic patients were included in those 16 enrolled subjects, but seven subjects were diagnosed as impaired glucose tolerance (IGT) (13) with no diabetic symptom or complication. HbAic and plasma glucose concentrations before and 2 hr after OGTT in those IGT subjects were 5.2% ± 0.3%, 5.4 ± 0.2 mM, and 8.4 ± 0.6 mM, respectively.

The ethics committee of Oita Medical University authorized the present study. All patients gave written informed consent to participate in the study according to Helsinki Declaration II.

VLCD-CJ Therapy. As an acclimatization control period, each subject was provided with a weight-maintenance diet of 1600 to 2000 kcal/day matched for each individual for 5 days prior to initiating a weight reduction program. All 16 subjects were introduced into very-low-calorie conventional Japanese diet (VLCD-CJ) therapy without any scheduled exercise (14). According to the program, the energy intake reduced to 1200, 1000, and 700 kcal/day at 3-day intervals. Succeeding these diets, a 380-kcal/day VLCD-CJ was provided for 14 days. After completion of the VLCD-CJ, energy intake was increased to 700, 1000, and 1200 kcal/day at 3-day intervals so that the subjects could maintain a reduced diet after discharge. To confirm weight loss of more than 10% compared with initial body weight, the VLCD-CJ was prolonged up to 2 additional weeks. The average time of VLCD-CJ therapy was 39.9 ± 1.8 days. VLCD-CJ consisted of 38 g/day protein, 45 g/day carbohydrate, and less than 4.0 g/day fat, supplemented with vitamins and minerals necessary for recommended daily dosages. All the diets provided were served as a conventional Japanese diet comprised of fiber-rich and non- or low-energy food stuffs (14). The nutritional content in the VLCD-CJ was almost the same as that in the 360-kcal VLCD (15). During the course of the program, body weight was measured daily immediately after waking up in the morning for 10 days before and continuing after the weight reduction program was started. Reduction rate of BMI was expressed as a ratio of the difference between BMIs before and after VLCD therapy to the initial BMI.

Analysis of HRV. Twenty-four-hour ambulatory ECG was recorded using a two-channel tape recorder (Fukuda Denshi Co., Tokyo, Japan) three times: at the time of admission, on the 14th day of the VLCD-CJ, and on the final day of the weight reduction program. While hospitalized, each patient was provided with a daily schedule to keep physical activity constant without a specific exercise regimen. No significant effect of physical activity on HRV was confirmed by calorie counts. Data were analyzed with a Marquette 8000 scanner (Marquette Electronics, Inc., Milwaukee, WI), using the standard Marquette algorithms for QRS labeling and editing.

According to these data, a mean normal R-R interval (mNN) was calculated and the five time-domain indices were expressed as follows: standard deviation of all NN intervals in the whole recording (SDNN), standard deviation of the average of NN intervals every 5-min period (SDANN), mean of the standard deviations of all NN intervals every 5-min period (SDNN index), number of pairs of adjacent NN intervals differing by more than 50 msec divided by total number of the NN interval (pNN50), and the square root of the mean of the sum of the squares of differences between adjacent NN intervals (RMSSD).

Power spectrum for HRV was analyzed by the Fast Fourier Transform Algorithm expressed as a square root of the areas under the power spectrum. The power frequency was differentiated into low (LF, 0.04–0.15 Hz) and high (HF, 0.15–0.40 Hz) components so that HF and a ratio of LF to HF (LF/HF) were calculated over a 24-hr period. To examine daily fluctuations of HRV, the HF and LF/HF variation was analyzed at the following 3-hr periods from 0:00 to 3:00 hr, from 9:00 to 12:00 hr, and from 15:00 to 18:00 hr, when HRV parameters were ascertained to be unaffected (16, 17). Rate of change in these cardioautonomic parameters was defined as a ratio of difference from initial value to the corresponding initial value.

Echocardiographic Study. To assess the effects of weight reduction on cardiac function, each subject was examined twice: before initiation and after the completion of the weight reduction program, by an ultrasonograph equipped with a 2.5 and 3.75 MHz adjustable transducer (sonolayer α SSA-270A, Toshiba Co., Tokyo, Japan). Left ventricular end-diastolic (LVDd) and end-systolic dimensions (LVDs), and mitral valve diastolic descent rate (MVDDR) and isometric relaxation time (IRT) were measured. Left ventricular ejection fraction (LVEF) and the

ratio of preejection period to left ventricular ejection time (PEP/LVET) were calculated.

Statistical Analysis. The data were expressed as means ± SEM except as otherwise described. Wilcoxon test was used to evaluate the differences between the parameters before and after weight reduction. The correlation between percent changes in the HRV indices and the parameters of body weight change induced by the weight reduction program was assessed by means of Spearman's rank correlation test. A level of $P < 0.05$ was defined as statistically significant.

Results

Changes in Body Weight and BMI. Following the VLCD-CJ therapy, all 16 subjects reduced their body weight by an average of 17.8 ± 1.5% from their initial values ($P < 0.01$). In contrast, mean blood pressures did not change (Table I). There was a positive correlation between initial BMI and the reduction rate of BMI ($y = -0.13 + 0.01x$, $r = 0.72$, $P < 0.01$). No adverse cardiac event such as arrhythmia or hypotension was found in any subject either during or after the VLCD-CJ therapy.

Changes in NN Intervals and HRV. Effects of weight reduction by more than 10% following VLCD-CJ on mNN and HRV parameters are shown in Table II. The parameters mNN, SDNN, SDANN, and pNN50 were increased on the 14th day of the 360-kcal diet in the VLCD-CJ program ($P < 0.01$ for mNN and $P < 0.05$ for the remaining parameters). The mNN and the five time-domain indices were all increased after the completion of the program ($P < 0.01$ for each). Both HF and LF increased concomitantly from 12 ± 1 msec to 16 ± 2 msec and from 21 ± 2 msec to 24 ± 2 msec after the weight reduction program ($P < 0.01$ for each). In contrast, LF/HF decreased on the 14th day of the 360-kcal diet in the VLCD-CJ program and at the completion of the program ($P < 0.01$ for each). A typical illustration of changes in power frequency domain of HRV from a 57-year-old female is shown in Figure 1, which shows that the amplitude of the HF component increased from 10 to 15 msec as her BMI decreased from 32.3 to 25.4 kg/m^2.

Comparative assessment of daily fluctuation of HF and LF/HF components before and after VLCD-CJ, based on spectral component analysis of each 3-hr period, demonstrated that the HF component dominated throughout all three periods of 00:00 to 03:00 hr, 09:00 to 12:00 hr, and 15:00 to 18:00 hr after VLCD-CJ ($P < 0.05$ for each). Unlike the HF component, the LF/HF component weakened in the 00:00 to 03:00-hr period after VLCD-CJ ($P < 0.05$ for each; Fig. 2).

Table II. Changes in NN Intervals and Heart Rate Variability during the 24-hr Period Following VLCD-CJ Therapy in 16 Patients

Parameters	Before	During	After
mNN (msec)	805 ± 21	864 ± 31[a]	930 ± 22[b,c]
SDNN (msec)	143 ± 10	165 ± 16[a]	184 ± 9[b]
SDANN (msec)	131 ± 10	150 ± 16[a]	172 ± 8[b]
SDNN index (msec)	51 ± 4	57 ± 6	64 ± 4[b]
pNN50 (%)	7 ± 1	12 ± 2[a]	16 ± 3[b]
RMSSD (msec)	27 ± 2	32 ± 3	38 ± 4[b]
HF (msec)	12 ± 1	13 ± 1	16 ± 2[b,c]
LF/HF	1.9 ± 0.1	1.7 ± 0.1[a]	1.5 ± 0.1[b]

Note. Values, means ± SEM. Before and After, before and after VLCD-CJ therapy; During, on the 14th day of the VLCD-CJ; mNN, mean normal RR interval; SDNN, standard deviation of NN intervals; SDANN, standard deviation of the mean NN intervals every 5-min segment; SDNN index, mean of standard deviation of NN intervals every 5-min segments; pNN50, number of pairs of adjacent NN intervals differing by more than 50 msec divided by total number of NN intervals; RMSSD, square root of mean of the sum of the square of difference, between adjacent NN intervals.
[a] $P < 0.05$, [b] $P < 0.01$ vs the corresponding values before VLCD-CJ therapy. [c] $P < 0.05$ vs the corresponding values during VLCD-CJ therapy.

Correlation between BMI and Cardioautonomic Parameters. A series of correlation matrices between BMI and mNN and HRV obtained from 24-hr recordings following weight reduction are shown in Table III. There was no correlation between initial BMI and initial cardioautonomic parameters, including mNN and HRV, and no correlation between reduction rate of BMI and initial cardioautonomic parameters (data not shown). Initial BMI correlated positively with rates of change in mNN, time-domain parameters except for pNN50, and HF, but not with rate of change in LF/HF. By comparison with initial BMI, reduction rate of BMI correlated positively with rates of change in all the cardioautonomic parameters except for the LF/HF.

Changes in Cardiographic Parameters Following VLCD-CJ. Table IV shows the changes in cardiographic parameters before and after VLCD-CJ. IRT, a parameter of diastolic function, LVDd, LVDs, and PEP/LVET decreased after the VLCD-CJ ($P < 0.01$ for each), while

Table I. Changes in Body Weight, BMI, and Mean Blood Pressure Following VLCD-CJ Therapy

	Before	After	% Difference
Body weight (kg)	77.3 ± 4.0	62.9 ± 2.6[a]	17.8 ± 1.5
BMI (kg/m^2)	31.4 ± 1.1	25.6 ± 0.7[a]	17.8 ± 1.5
Mean blood pressure (mmHg)	91.6 ± 2.2	90.0 ± 2.1	1.2 ± 0.4

Note. Values, means ± SEM. Before and After, before and after VLCD-CJ therapy. % difference, % difference between values before and after VLCD-CJ/initial values.
[a] $P < 0.01$ vs the corresponding values before VLCD-CJ therapy.

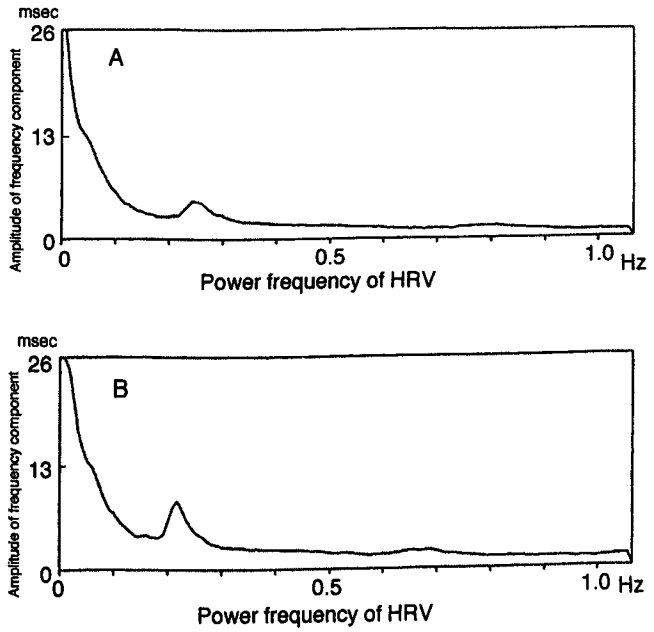

Figure 1. A typical illustration of changes in power frequency domain of HRV from a 57-year-old female with 35.5 kg/m^2 BMI reduced to 26.6 kg/m^2. (A) The spectral analysis during the baseline 24-hr period. (B) The analysis after weight reduction. Note that weight reduction increased the amplitude of a HF component (0.15–0.40 Hz) predominantly from 10 msec (A) to 15 msec (B).

LVEF, a parameter of systolic function, and MVDDR increased after the VLCD-CJ ($P < 0.01$ for each).

Discussion

To examine the effects of weight reduction on autonomic nerve activity in obesity, HRV was investigated using 24-hr ECG recordings. The present study has demonstrated several main findings. Mean body weight loss exceeded 10% after VLCD-CJ therapy. Under such weight reduction during a relatively short term, an HF component assessed by power spectrum was strengthened, but an LF/HF component was weakened, together with increases in RR interval and five time-domain indices. This association of HF dominance with LF/HF suppression fluctuated daily, but the pattern strengthened, particularly during mid-night (00:00–03:00 hr), after weight reduction over 10%. The change in HF, but not LF/HF, was significantly correlated with weight reduction. These results lead us to conclude that weight reduction of more than 10% after VLCD-CJ therapy improves parasympathetic activity in obese patients. The findings were derived from an open trial study because ethical limitations did not permit us to add an obese control group without diet treatment.

Changes in cardiac vagal activity in response to cardiovascular reflex tests indicated lower parasympathetic and higher sympathetic activity in obese patients (18). A microneurographic study showed higher sympathetic activity innervating the muscle in the obese (19). A study of HRV and cardioautonomic changes following weight reduction showed that 10% weight loss failed to improve the lowered parasympathetic activity in obese subjects (9). In contrast, a parasympathetic and sympathetic blocker study with sequential and parenteral administration of antagonists showed that obese subjects increased parasympathetic activity and decreased sympathetic activity after the same weight loss using evaluation of mean RR interval change (10). The RR interval is thus influenced by autonomic nerve activity. It is generally accepted that parasympathetic activity is the major contributor to the HF value obtained from power spectrum analysis (20). On the other hand, agreement on the LF or LF/HF component as a marker of sympathetic modulation is not generally accepted (21). A recent report using HRV power spectrum analysis showed that parasympathetic activity was restored to normal when the subjects were compelled to reduce their body weight quite rapidly by gastroplasty (22). Our current results show that the lowered

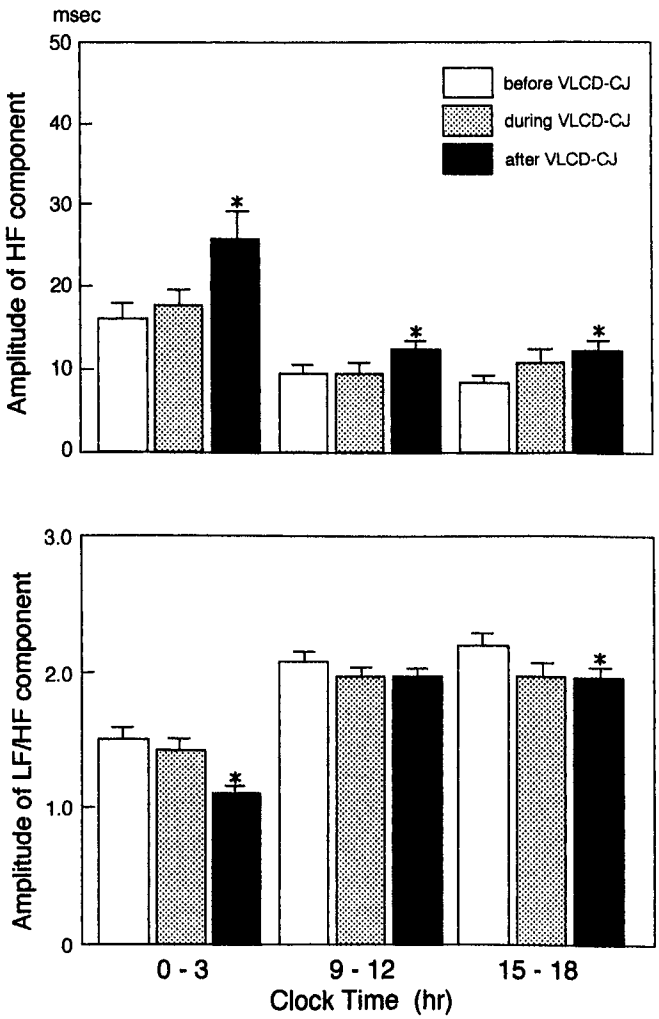

Figure 2. The effects of weight reduction on daily amplitude fluctuation of each component. The amplitudes were shown at every 3-hr period, but those at three meals of VLCD-CJ were not measured to exclude effects of postprandial parasympathetic activation. Note that HF component dominated throughout the whole three periods after VLCD-CJ therapy, but the ratio of LF/HF components were solely suppressed in the midnight of the 00:00- to 03:00-hr period. During, on the 14th day of VLCD-CJ. After, at the completion of the weight reduction program. * $P < 0.05$ vs the corresponding values before VLCD-CJ therapy.

Table III. Correlation Matrices Between BMI and Cardioautonomic Parameters

Parameters/ rate of change in	Initial BMI		Reduction rate of BMI	
	Coefficient value	*P* value	Coefficient value	*P* value
mNN	0.693	0.003	0.538	0.032
SDNN	0.676	0.004	0.750	0.001
SDANN	0.684	0.004	0.713	0.002
SDNN index	0.651	0.006	0.718	0.002
RMSSD	0.603	0.013	0.610	0.012
pNN50	0.423	NS	0.573	0.021
HF	0.562	0.023	0.623	0.010
LF/HF	0.126	NS	−0.083	NS

Note. Reduction rate of BMI and rate of change in cardioautonomic parameters, difference from initial value/the corresponding initial value. NS, not significant.

HF component, a marker of parasympathetic activity, was much improved in response to a smaller weight reduction during a shorter period. This is consistent with previous findings that effective weight reduction influenced parasympathetic activity in the obese independently of usage of HRV parameters. The weakened LF/HF component after over 10% weight loss revealed in the present study also corresponded with the results of the blocker study (10). On the other hand, the present study demonstrates that improvement of the HF component was significantly correlated with reduction of BMI, whereas the LF/HF component was not. This finding indicates that the HF component is more reliable for assessment of cardioautonomic activity affected by weight reduction.

The present long-term recording of cardioautonomic activity enables us to analyze changes in daily fluctuation. Consistent with previous findings (19), HF and LF/HF components in the present study exhibited daily fluctuations, but also exhibited a reciprocal pattern of greater sympathetic activity in the daytime and greater parasympathetic activity at night. Changes in both HF and LF/HF after weight reduction were greater at night. However, HF changes were greater than those of LF/HF throughout the measured period. In other words, the evaluation of HF and LF/HF fluctuations in obese subjects suggests a characteristic alteration of autonomic nerve activity following weight reduction. These findings, in turn, lead us to assume that diurnal rhythm of autonomic nerve activity is attenuated in obese patients. Indeed, distortion or disappearance of circadian rhythm is detectable in feeding behavior, as well as energy metabolism in obese animals (12).

A critical question can be raised as to how parasympathetic activity is enhanced in response to the reduced BMI. There is no definitive answer to the question. The magnitude of changes in the HRV parameters after completion of the weight reduction program was greater than that during the VLCD-CJ regimen of 380 kcal/day. For this reason it seems quite unlikely that hormonal and metabolic factors resulting from negative energy balance induced by the VLCD-CJ therapy are directly involved in the improvement. One possible explanation is the effect of hemodynamic abnormalities associated with obesity. Increases in cardiac output and blood volume and lowered total peripheral vascular resistance are well-known features of obese subjects. To compensate for increased oxygen consumption, development of obesity elevates both blood volume and cardiac output (23). Myocardial contractility is concomitantly diminished in obesity (24). In fact, systolic function assessed by LVEF was significantly improved after weight reduction in the present study, suggesting the presence of impaired cardiac performance, even in asymptomatic obesity. Reversibility of the impaired cardiac function following weight reduction has been reported to depend on the degree and duration of obesity, as well as fat distribution (25). Taken together, it seems likely that increased parasympathetic activity after the present weight reduction mediates the improvement of cardiac functions shown in the present study. Another possible explanation is that loss of visceral and thoracic fat causes functional improvement of both pulmonary residual and expiratory reserve volume. The increase in vital capacity that is shown to correlate positively with decrease in BMI (26) simultaneously induces activation of parasympathetic nerves through stimulation of lung stretch receptors. Respiratory frequency *per se* has been shown to affect the HF component independently of modulation of parasympathetic activity (27). However, parasympathetic measures made after drug blockade, which is not greatly affected by respiration, showed increased parasympathetic activity after weight reduction (10). Based on these observations, it seems reasonable to submit that vital capacity significantly affected parasympathetic activity in the present study, whereas respiratory frequency may not be a significant factor during weight reduction.

In contrast to human obesity, pathogenesis in animal obesity models has been explained partly by impairment of the autonomic nervous system, *i.e.*, an increase in parasympathetic nerve activity and a relative decrease in sympa-

Table IV. Changes in Cardiographic Parameters Following Weight Reduction

	LVDd(mm)	LVDs(mm)	LVEF(%)	MVDDR(mm)	PEP/ILVET	IRT(msec)
Before	50.0 ± 1.4	33.9 ± 1.7	63.7 ± 2.2	65.3 ± 5.6	0.393 ± 0.017	87.3 ± 2.4
After	45.7 ± 1.0[a]	28.4 ± 1.2[a]	72.3 ± 1.7[a]	99.1 ± 9.2[a]	0.361 ± 0.012[a]	72.0 ± 2.2[a]

Note. Values, means ± SEM. LVDd, left ventricular dimensions in diastole. LVDs, left ventricular dimensions in systole. LVEF, left ventricular ejection fraction. MVDDR, mitral valve diastolic descent rate. PEP/LVET, ratio of preejection period to left ventricular ejection time. IRT, isometric relaxation time. Before, before VLCD-CJ therapy. After, after VLCD-CJ therapy.
[a] $P < 0.01$ vs the corresponding values before VLCD-CJ therapy.

thetic activity (28–30). Of note, regional sympathetic nerve activity is heterogeneous in mammals. Sympathetic nerve activity in the kidneys is elevated, while that in the heart is suppressed in obesity (31). The accelerated cardiac sympathetic function shown in the present obese subjects may be attributed to the maintenance of energy homeostasis under such increases in energy demand. It is worth noting that the current findings based on HRV measures have to be restricted to cardiovascular function because the abnormality in sympatho-vagal activity in the steady-state obese subjects and its improvement after weight reduction may be totally different in different organs.

In conclusion, the present study has demonstrated that short-term weight reduction over 10% improves parameters related to HRV in obese subjects, particularly an increase in the HF component, an indicator of parasympathetic activity, throughout day and night periods. From the viewpoint of the pathogenesis of lowered parasympathetic activity, this improvement may mediate simultaneous reduction of risks such as life threatening arrhythmia and sudden death in obese subjects.

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