

Autonomic Nervous System Activity and the State and Development of Obesity in Japanese School Children

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Abstract

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Objective: The autonomic nervous system (ANS) plays an important role in regulating energy expenditure and body fat content; however, the extent to which the ANS contributes to pediatric obesity remains inconclusive. The aim of this study was to evaluate whether sympathetic and/or the parasympathetic nerve activities were altered in an obese pediatric population. We further examined a physiological association between the duration of obesity and the sympatho-vagal activities to scrutinize the nature of ANS alteration as a possible etiologic factor of childhood obesity.

Research Methods and Procedures: Forty-two obese and 42 non-obese healthy sedentary school children were carefully selected from 1080 participants initially recruited to this study. The two groups were matched in age, gender, and height. The clinical records of physical characteristics and development of the obese children were retrospectively reviewed to investigate the onset and progression of obesity. The ANS activities were assessed during a resting condition by means of heart rate variability power spectral analysis, which enables us to identify separate frequency components, i.e., total power (TP), low-frequency (LF) power, and high-frequency (HF) power. The spectral powers were then logarithmically transformed for statistical testing.

Results: The obese children demonstrated a significantly lower TP (6.77 ± 0.12 vs. 7.11 ± 0.04 ln ms^2 , $p < 0.05$), LF power (6.16 ± 0.12 vs. 6.42 ± 0.05 ln ms^2 , $p < 0.05$), and HF power (5.84 ± 0.15 vs. 6.34 ± 0.07 ln ms^2 , $p < 0.01$) compared with the non-obese children. A partial correlation analysis revealed that the LF and HF powers among 42 obese children were negatively associated with the duration of obesity independent of age (LF: partial $r = -0.55$, $p < 0.001$; HF: partial $r = -0.40$, $p < 0.01$). The obese children were further subdivided into two groups based on the length of their obesity. All three spectral powers were significantly reduced in the obese group with obesity of >3 years ($n = 18$) compared to the group with obesity of <3 years.

Discussion: Our data indicate that obese children possess reduced sympathetic as well as parasympathetic nerve activities. Such autonomic depression, which is associated with the duration of obesity, could be a physiological factor promoting the state and development of obesity. These findings further imply that preventing and treating obesity beginning in the childhood years could be an urgent and crucial pediatric public health issue.

Key words: childhood obesity, duration of obesity, sympathetic nervous system, parasympathetic nervous system, heart rate variability

Introduction

An increase in obesity prevalence has been observed internationally in children from preschool age to adolescence (1). Alarming findings are now emerging from Asian countries that are rapidly westernizing their lifestyles, particularly their behavioral and dietary habits. As for Japan, the 2000 annual report of the health and physical development of the pediatric population disclosed that the number of obese children ages 6 to 11 years has approximately doubled over the last 20 years (2). Data from previous studies provide strong evidence that a higher level of body

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mass index (BMI)¹ during childhood can predict overweight later in life (3,4). To make matters worse, obesity in children is associated with various adverse biochemical, physiological, and psychological effects, many of which have strong links with chronic disease risk factors in adulthood (4,5). Thus, finding etiologic factors of obesity, as well as preventing the obesity epidemic beginning in the childhood years, is currently a critical issue in the pediatric public health research field.

Although many environmental and genetic factors are intermingled, the development of obesity is ultimately caused by a sequential alteration of energy balance, with energy intake exceeding energy expenditure (6). The stability of a human's internal environment depends largely on the orchestrations of the autonomic nervous system (ANS). Because the sympathetic branch of the system particularly contributes to coordinating energy homeostasis, the alteration of sympathetic nervous system (SNS) activity is widely assumed to promote onset and development of obesity. The MONA LISA hypothesis, an acronym for *Most Obesities kNown Are Low In Sympathetic Activity* (7), has been supported (8,9). However, disagreement still exists over the nature of the sympathetic abnormality within the adult obese population (10,11). Less research has been done with children, and the findings regarding the physiological role of the SNS on pediatric obesity have been thus far inconclusive (12–14). The discrepancy is thought to arise largely from the difficulty in controlling the array of variables (including age, gender, the history of obesity, other medical complications, dietary and behavioral habits, physical activity levels, and emotional stress) and in adequately assessing SNS activity in human subjects of all age groups.

Heart rate variability (HRV) power spectral analysis is a well-accepted, useful, and noninvasive method, and has provided a comprehensive quantitative and qualitative evaluation of neuroautonomic function under various research and clinical settings (15–17). In general, power spectral analysis of HRV has shown at least two distinct regions of periodicity in electrocardiogram (ECG) R-R intervals. The high-frequency component (>0.15 Hz) is a major contributor to reflecting parasympathetic nervous system (PNS) activity, and the low-frequency component (<0.15 Hz) is associated with both SNS and the PNS activities (18,19). Previous investigations have demonstrated that the percentage of body fat (20), changes in body weight and energy storage (21), and glucose-induced thermogenesis (22) were correlated with differences in the power spectral components. A series of our recent studies with the HRV power

spectral analysis have shown that obese young women possess significantly lower SNS activity against various thermogenic perturbations, such as cold exposure (23), capsaicin-containing yellow curry diet (24), and mixed food intake (25). Unlike invasive measurements such as plasma catecholamine concentration, catecholamine turnover, and muscle sympathetic nerve activity, the HRV power spectral analysis lightens the burden imposed on subjects during an experiment and is a suitable and valuable approach to evaluating ANS activity in large-scale pediatric obesity research.

Accordingly, in this study, we evaluated resting ANS activity by means of the HRV power spectral analysis in healthy sedentary non-obese and obese school children who were carefully selected to avoid the physiological heterogeneity of obesity, and investigated whether the SNS and/or the PNS activities were altered in an obese pediatric population. We also retrospectively reviewed clinical records of physical characteristics and development within the obese children, and examined a physiological association between the history of obesity (the onset and progression of obesity) and the sympathovagal activities in order to further scrutinize the nature of ANS alteration as a possible etiologic factor of childhood obesity.

Research Methods and Procedures

Subjects

One thousand eighty obese and non-obese healthy Japanese children, ages 6 to 12 years, initially volunteered to participate in this research. The study protocol was approved by the Institutional Review Board of Kyoto University Graduate School. All children and their parents were carefully instructed about the study and gave their written informed consent to participate in the study. Before obtaining any data from the children, the parents completed a standardized health questionnaire regarding their children's past medical history, current health condition, past records of height and body weight for estimating the history of obesity, diet, physical activity, and lifestyle.

After measuring height and body weight, percentage of body fat was determined by means of a bioelectrical impedance analyzer (Model TBF-534; Tanita Corp., Tokyo, Japan). The analyzer has been used in several pediatric investigations because it produces a reasonable estimate for body fat content in children (26,27). BMI was also calculated as body weight divided by square height. "Obesity" was defined based on the criterion previously used in pediatric research (>120% of the standard body weight for Japanese children) (3).

The results of the health questionnaires and the body composition measurements were carefully examined. Then, 42 obese and 42 non-obese children were selected for investigation of a physiological association of childhood

¹ Nonstandard abbreviations: BMI, body mass index; ANS, autonomic nervous system; SNS, sympathetic nervous system; HRV, heart rate variability; ECG, electrocardiogram; PNS, parasympathetic nervous system; TP, total power; LF, low frequency; HF, high frequency; MSNA, muscle sympathetic nerve activity.

Table 1. Physical characteristics of children

	Obese (n = 42)	Non-obese (n = 42)
Age (years)	9.0 ± 0.3	9.0 ± 0.3
Gender (boy/girl)	18/24	18/24
Height (cm)	133.3 ± 2.1	135.2 ± 1.8
Body weight (kg)	43.6 ± 1.9*	31.8 ± 1.4
BMI (kg/m ²)	23.4 ± 0.5*	17.4 ± 0.3
Percentage of standard body weight (%)	136.4 ± 2.1*	100.9 ± 1.2
Percentage body fat (%)	29.5 ± 0.7*	17.9 ± 0.6

Values are expressed as means ± SE.

* $p < 0.001$.

obesity and ANS activity. The two groups were matched in age, gender, and height. All children were in good health and had no personal or family history of hypertension, cardiovascular disease, diabetes mellitus, or other endocrine diseases. None of the children were taking any medications. According to the nutritional survey included in the health questionnaire, daily energy intake and nutritional content of food did not significantly differ between the two groups. Concerning the physical activity level, none of the children in these groups regularly engaged in sports activities or aerobic exercises. The descriptive characteristics of the children are presented in Table 1.

Methods

The children came at 8:30 AM to the temporary laboratory that was set up in the school infirmary. All experiments were performed in the morning, and the entire research project lasted for 2 consecutive days in June. The room was temperature controlled (25 °C), quiet, and comfortable, with minimal arousal stimuli. After appropriate skin preparation, the subjects were fitted with ECG electrodes and then rested for at least 15 minutes before the start of the experiment.

After the resting period, the CM₅ lead ECG signals were continuously recorded for 4.5 minutes while each child remained seated in a chair and breathed normally (28). It should be noted that, according to our preliminary experiment as well as accepted parameters of pediatric physiology (29), children's breathing frequency is generally higher than 9 rates per minute (>0.15 Hz). Thus, we assumed that, without controlling the respiration rates during the ECG measurements, respiratory-linked variations in heart rate did not overlap with low-frequency heart rate fluctuations (<0.15 Hz) from other sources.

R-R Spectral Analysis Procedure

Our R-R interval power spectral analysis procedures have been fully described elsewhere (16,30,31). Briefly, analog output of the ECG monitor (MEG-6100; Nihon Kohden, Tokyo, Japan) was digitized through a 13-bit analog-to-digital converter (HTB 410; Trans Era, South Orem, UT) at a sampling rate of 1024 Hz. The digitized ECG signals were differentiated, and the resultant ECG QRS spikes and the intervals of the impulses (R-R intervals) were stored sequentially on a hard disk for later analyses.

Before R-R spectral analysis was performed, the stored R-R interval data were displayed and aligned sequentially to obtain equally spaced samples with an effective sampling frequency of 2 Hz (32) and displayed on a computer screen for visual inspection. Then, the direct current component and linear trend were completely eliminated by digital filtering for the band-pass between 0.03 and 0.5 Hz. The root mean square value of the R-R interval was calculated as representing the average amplitude. After passing through the Hamming-type data window, power spectral analysis by means of a fast Fourier transform was performed on a consecutive 256-s time series of R-R interval data obtained during the test.

Based on our previous investigations (16,30,31,33), the spectral powers in frequency domain were quantified by integrating the areas under the curves for the following respective band width: the low-frequency (LF: 0.03 and 0.15 Hz), an indicator of both SNS and PNS activity; the high-frequency (HF: 0.15 and 0.5 Hz), which solely reflects the PNS activity; and the total power (TP: 0.03 and 0.5 Hz), representing the overall ANS activity. Because basal spectral absolute values differed greatly among individuals, the spectral powers were then logarithmically transformed for statistical testing (17–19).

Statistical Analyses

All data are expressed as mean ± SE. Student's unpaired *t* test was performed to assess statistical differences in physical characteristics and the parameters of the ANS activity between the obese and non-obese groups. Partial correlation analysis was performed to examine the relationships between the ANS parameters and the duration of obesity independently of a covariate, i.e., age. All *p* values were two-sided. A *p* value of <0.05 was chosen as the level of significance. All statistical analyses were made on a personal computer with a commercial software package (SPSS version 10.0J for Windows; SPSS Inc., Chicago, IL).

Results

Figure 1 represents typical sets of raw R-R intervals and the power spectral data obtained from an obese and a non-obese child, respectively, during the resting condition.

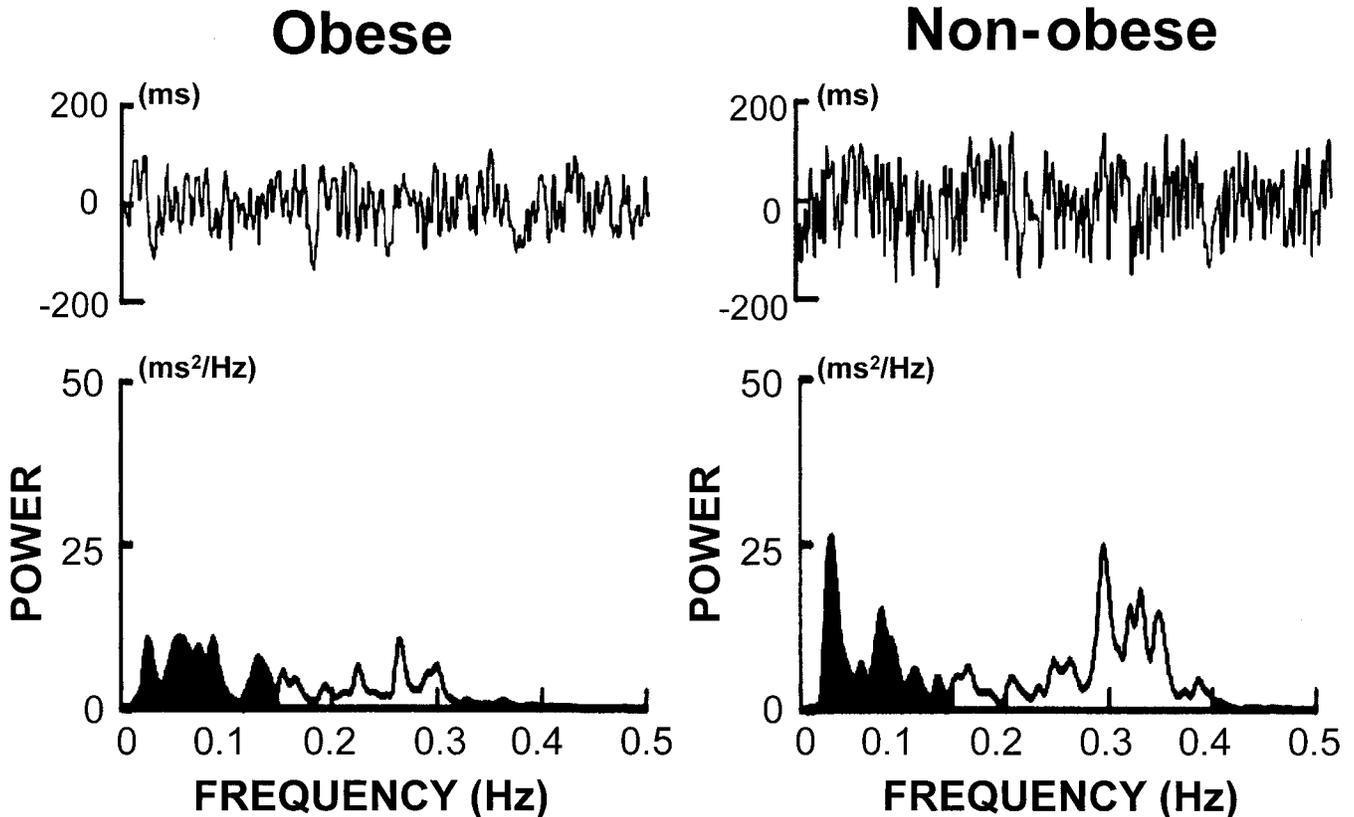


Figure 1: Examples of ECG R-R interval changes and the corresponding power spectra for an obese and a non-obese child at rest, respectively.

According to visual inspection, the obese child possessed remarkably reduced ranges in R-R variability, as well as in both frequency components of the power spectrum, compared with the non-obese child.

The statistical analysis revealed that there were significant differences in the R-R spectral parameters between the two groups. The obese group had a significantly higher resting heart rate compared with the non-obese group (90.7 ± 1.5 vs. 84.3 ± 1.0 beats per minute, $p < 0.001$). As Figure 2 demonstrates, all the spectral powers were significantly lower in the obese group than in the non-obese group (TP: 6.77 ± 0.12 vs. 7.11 ± 0.04 $\ln \text{ms}^2$, $p < 0.05$; LF: 6.16 ± 0.12 vs. 6.42 ± 0.05 $\ln \text{ms}^2$, $p < 0.05$; HF: 5.84 ± 0.15 vs. 6.34 ± 0.07 $\ln \text{ms}^2$, $p < 0.01$), indicating that both SNS and PNS activity decrease in obese children.

To further investigate an association between the HRV spectral powers and the history of obesity, the obese children were subdivided into two groups based on the length of obesity. It has been shown that physiological factors such as age influence HRV (34,35); thus, we performed the partial correlation analysis to examine the relationship after adjustment for age. As Figure 3 shows, the LF and HF powers among 42 obese children were negatively correlated with the duration of obesity (LF: partial $r = -0.55$, $p < 0.001$,

HF: partial $r = -0.40$, $p < 0.01$). A statistical analysis demonstrated that all three spectral powers were significantly reduced in the group with obesity of >3 years ($n = 18$) compared to the group with obesity of <3 years ($n = 24$; TP: 6.35 ± 0.15 vs. 7.33 ± 0.09 $\ln \text{ms}^2$, $p < 0.01$; LF: 5.78 ± 0.15 vs. 6.67 ± 0.12 $\ln \text{ms}^2$, $p < 0.01$; HF: 5.37 ± 0.20 vs. 6.46 ± 0.13 $\ln \text{ms}^2$, $p < 0.01$).

Discussion

This study provides valuable information regarding a potential etiologic association between childhood obesity and ANS activity. The main findings are that healthy sedentary obese school children possess much lower sympathetic and parasympathetic nerve activities. In addition, a degree of these autonomic reductions depends on the duration of obesity independent of a subject's age.

It is well known that the coordination of energy homeostasis particularly relies on the normal functioning of the sympathoadrenal system. As the MONA LISA hypothesis indicates (7), it is reasonable to assume that reduced SNS activity leads to a lower rate of thermogenesis, and consequently, to a positive energy balance and obesity. Despite intensive research, physiological roles of SNS ac-

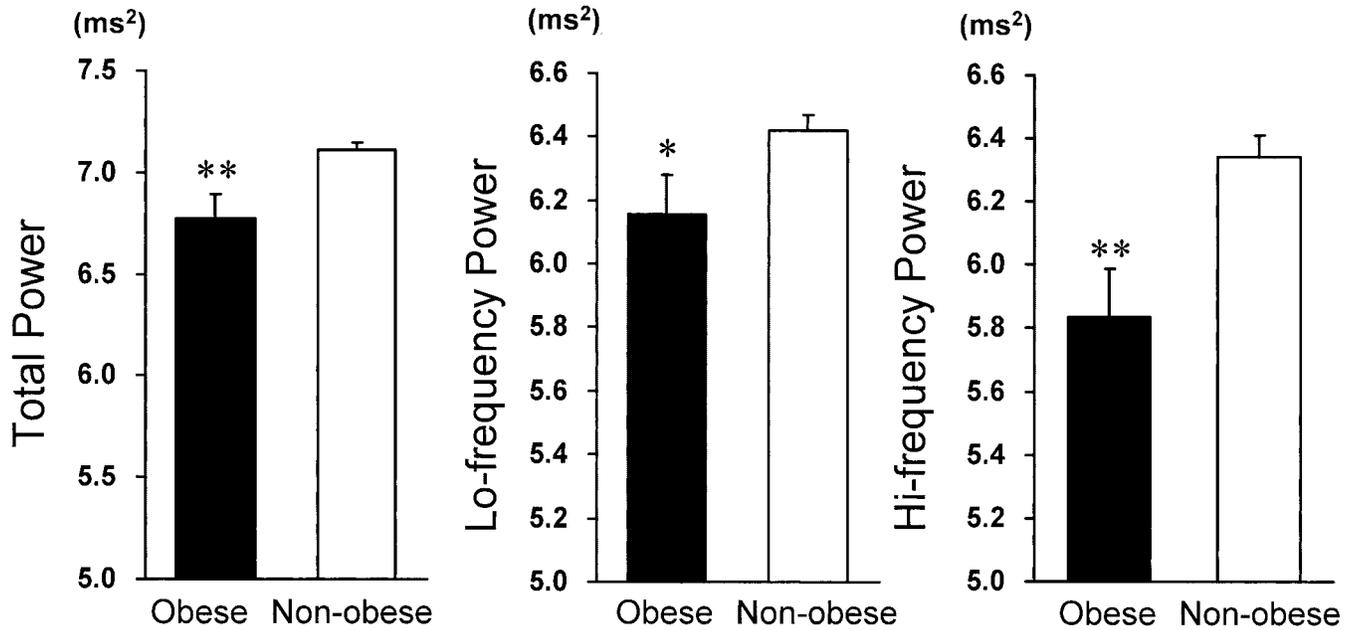


Figure 2: Comparison of total, low-frequency, and high-frequency powers between the obese and the non-obese groups, respectively. Results are expressed as mean \pm SE for each group. * $p < 0.05$, ** $p < 0.01$.

tivity on human obesity remain unclear. The different methodologies used among investigators may have caused this situation. For instance, plasma and urinary norepinephrine estimates, both of which are commonly used as global indexes for sympathetic activity, have provided conflicting

results over the nature of the SNS affected by obesity (36). Technical developments have allowed direct intraneural recordings of sympathetic nerve traffic by microneurography, but the technique is limited to peripherally measuring SNS activity in the skin and skeletal muscles (9,37). The muscle

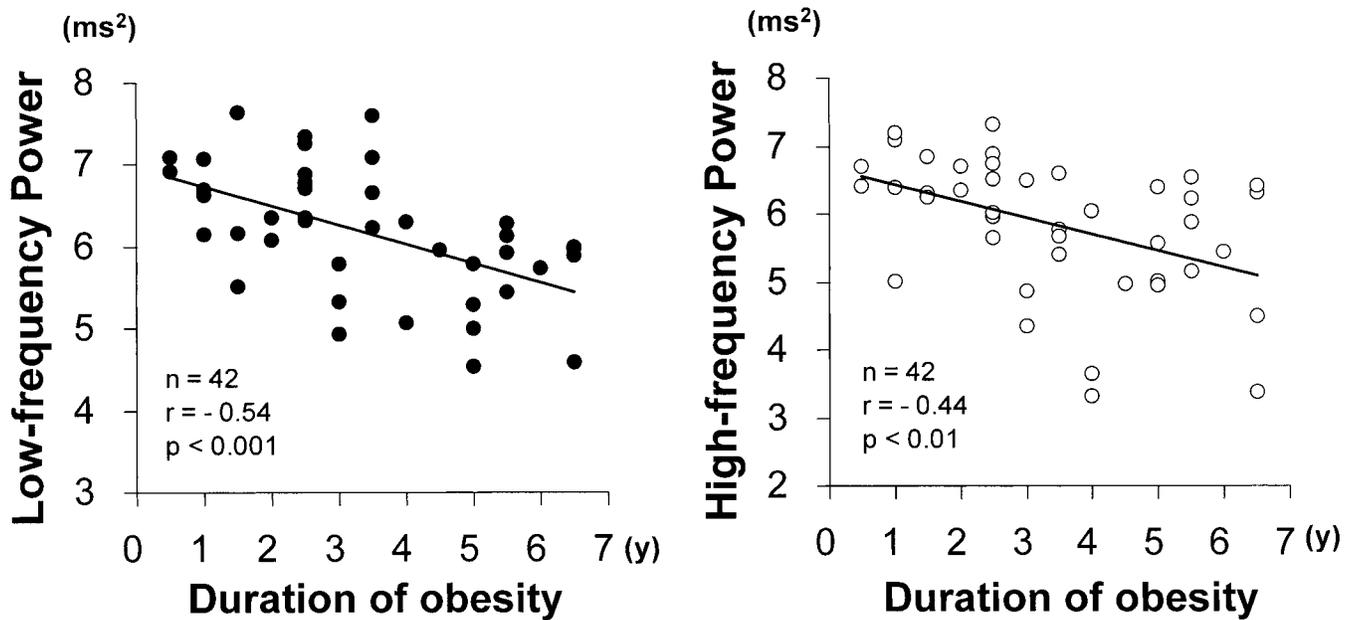


Figure 3: Partial correlations between both the low-frequency and the high-frequency powers and the duration of obesity, independent of a physiological effect of age among 42 obese children.

sympathetic nerve activity (MSNA) has been applied to obesity research, generally demonstrating a positive correlation between the amount of body fat and MSNA (10,11). Because measuring MSNA requires the insertion of a fine tungsten microelectrode and because MSNA is mainly related to the control of blood pressure rather than energy metabolism, the methodology is not relevant to investigating SNS activity, especially in a pediatric and epidemiological obesity study.

The spectral analysis of HRV has attracted considerable attention from investigators in various physiological fields, including diabetes and obesity research both for adult (16, 20–22, 31) and pediatric populations (13,14,38–41), and has become a reliable noninvasive measure to appraise ANS activity. As to an association of the ANS and energy homeostasis, Landsberg and Young (42) indicated that catecholamine turnover within cardiac tissue could precisely reflect autonomic events that affect energy metabolism elsewhere in the body. We have conducted a pharmacological blockade experiment to confirm the validity of the HRV power spectral analysis for evaluating and quantifying the sympathovagal activity. In short, after atropine, a parasympathetic muscarinic antagonist, was intravenously injected, R-R variability was markedly reduced and the HF components were almost entirely abolished, whereas the LF components were partly decreased. When propranolol, a β -adrenoceptor antagonist, was additionally injected, heart rate fluctuations were almost entirely abolished, and the resting energy expenditure was significantly reduced (23,25,31). Previous clinical studies further demonstrated that noninsulin-dependent diabetes mellitus patients with neuropathy had markedly reduced R-R interval fluctuations and HRV spectral power at rest compared with healthy individuals (16,30). These findings support the previous studies (18,19) and indicate that 1) the HF power is associated solely with PNS activity, and the LF power is jointly mediated by PNS and SNS activities, and 2) R-R interval variability and the integrated values of all the components of power spectra could reflect the overall ANS activity. Concerning the experimental setting, several studies have faced a difficulty in using HRV spectral analysis during nonresting conditions such as dynamic exercise, but the validity of spectral analysis of HRV under the resting condition, which we used in this study, has been well documented (16,17,43). Therefore, the application of HRV spectral analysis in a childhood population has distinct advantages over the existing methods for autonomic function in that it is noninvasive, less time consuming, and less upsetting for young participants during an experiment.

Although quantification and interpretation of HRV remain a complex issue (15,44), the efficacy and applicability of the technique used in the present investigation have been shown in previous research (16,30,31,33,45,46). By applying this analysis procedure, we measured the absolute val-

ues of spectral components to examine the ANS features of pediatric obesity, and provided the evidence that the TP, LF, and HF powers were all more substantially reduced in the obese than in the non-obese children. It has been pointed out that the frequency component of HRV spectral analysis, which reflects only the SNS activity, is difficult to single out. The normalized units and the ratios, i.e., LF/HF, have been used as alternative indexes to evaluate SNS and PNS activities (13,16,22). These indexes have been valuable and applicable to clinical settings as well as basic physiological research, but the results could overestimate or underestimate the sympatho-vagal activity because the indexes are relative values. As described above, neurophysiological contributions to the LF and HF powers have been clarified, and SNS activity, at least in part, does contribute to the LF power even in the resting condition (23). We recognize that the LF power, like all other indicators, has its limitations in precisely measuring SNS activity. Nevertheless, under the condition that the LF power together with the HF power and the TP all significantly decrease, which has appeared in presently healthy obese children, it is plausible to assume that both SNS and PNS activity could be reduced.

Even during childhood, low levels of PNS activity are associated with cardiac autonomic neuropathy in diabetics with poor metabolic control (39), duration of diabetes (40), and elevated blood pressure (47). Taking the previous results into consideration, reduced PNS activity appearing in presently healthy obese children might be a conceivable early sign to predict cardiovascular and metabolic health. As to the SNS activity, our results suggest the possibility that the MONA LISA hypothesis, indicating that obesity is associated with a relative or absolute reduction in the thermogenic component of SNS activity, might, at least in part, contribute to our understanding of the pathophysiological features of pediatric obesity. Normal or increased levels of SNS activity and its blunted responsiveness seem to be present in adult-established obesity (9,11,23–25). Taken together, the nature of SNS alteration in human obesity might diversify depending on the time in which the obese state occurs, is promoted, or is established.

We have extensively reviewed the literature regarding pediatric obesity. To the best of our knowledge, however, a limited number of studies have been conducted to examine the physiological role of ANS in human obesity during the early stage of life. Martini et al. (13) and Riva et al. (14) used time- and frequency-domain of HRV measured by 24-hour Holter recordings, and suggested that obese adolescents with metabolic changes, such as hyperinsulinemia, euglycemia, or dislipidemia, may have a sympathovagal balance, characterized by a primary decrease in PNS activity with a relative prevalence of SNS activity. Yakinci et al. (12) performed noninvasive autonomic tests, including orthostatic test, Valsalva maneuver, and deep breathing, and

indicated normal activity of SNS and hypoactivity of PNS in obese children. According to the study of Wawryk et al. (41), both LF and HF components were significantly reduced, which suggests autonomic suppression, in children with diabetes mellitus and greater body weight compared with the control group. Because of the disparity in the experimental conditions, i.e., subjects' age and clinical characteristics, the number of participants, and analytical procedures for the ANS activity, the outcomes of these investigations and our study were not always consistent. We assume, however, that the autonomic alteration is evident and might be an important etiologic factor of childhood obesity.

Other intriguing results obtained from this study were that the LF and HF powers decreased with increasing duration of obesity and that the depression of these powers was more apparent in the children with obesity of more than 3 years. Age has been reported to have a strong influence on HRV (34,35). Thus, we performed a partial correlation analysis, revealing that the reduction of the spectral powers was not related to a physiological effect of age. Future longitudinal research will be needed to confirm the changes of ANS activity and the development of obesity in the childhood. Nonetheless, the present data indicate that an increasing duration of obesity could affect the global levels and/or functions of SNS as well as PNS in simple obese children.

In summary, we investigated a potential physiological association of ANS activity and the state and development of obesity in healthy sedentary school children. Causes and consequences of human obesity continue to elude. Our findings, however, indicate that childhood obesity is closely interrelated with the reduction of both SNS and PNS activities. Because the ANS is involved in nearly every important homeostatic process going on within the body, the suppression of autonomic functioning can cause far-reaching adverse effects, including metabolic disorder and cardiovascular malfunction, and consequently, undermine children's health. In addition, the risk of obesity in adulthood is increased for obese children. Thus, this study further implies that preventing and treating obesity beginning in the childhood years is an urgent and crucial pediatric public health issue.

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