Quantifying cardiac sympathetic and parasympathetic nervous activities using principal dynamic modes analysis of heart rate variability

Yuru Zhong, Kung-Ming Jan, Ki Hwan Ju, and Ki H. Chon
1Department of Biomedical Engineering, State University of New York at Stony Brook, Stony Brook, New York; and 2Department of Cardiology, Columbia University, New York, New York

Submitted 3 January 2006; accepted in final form 27 March 2006

Zhong, Yuru, Kung-Ming Jan, Ki Hwan Ju, and Ki H. Chon. Quantifying cardiac sympathetic and parasympathetic nervous activities using principal dynamic modes analysis of heart rate variability. Am J Physiol Heart Circ Physiol 291: H1475–H1483, 2006. First published April 7, 2006; doi:10.1152/ajpheart.00005.2006.—The ratio between low-frequency (LF) and high-frequency (HF) spectral power of heart rate has been used as an approximate index for determining the autonomic nervous system (ANS) balance. An accurate assessment of the ANS balance can only be achieved if clear separation of the dynamics of the sympathetic and parasympathetic nervous activities can be obtained, which is a daunting task because they are nonlinear and have overlapping dynamics. In this study, a promising nonlinear method, termed the principal dynamic mode (PDM) method, is used to separate dynamic components of the sympathetic and parasympathetic nervous activities on the basis of the ECG signal, and the results are compared with the power spectral approach to assessing the ANS balance. The PDM analysis based on the 28 subjects consistently resulted in a clear separation of the two nervous systems, which have similar frequency characteristics for parasympathetic and sympathetic activities as those reported in the literature. With the application of atropine, in 13 of 15 supine subjects there was an increase in the sympathetic-to-parasympathetic ratio (SPR) due to a greater decrease of parasympathetic than sympathetic activity (P = 0.003), and all 13 subjects in the upright position had a decrease in SPR due to a greater decrease of sympathetic than parasympathetic activity (P < 0.001) with the application of propranolol. The LF-to-HF ratio calculated by the power spectral density is less accurate than the PDM because it is not able to separate the dynamics of the sympathetic and parasympathetic nervous systems. The culprit is equivalent decreases in both the sympathetic and parasympathetic activities irrespective of the pharmacological blockades. These findings suggest that the PDM shows promise as a noninvasive and quantitative marker of ANS imbalance, which has been shown to be a factor in many cardiac and stress-related diseases.

THE AUTONOMIC NERVOUS SYSTEM (ANS) consists of two counteracting subsystems: sympathetic (SNS) and parasympathetic nervous systems (PNS). Interactions between the SNS and PNS are essential for maintaining more efficient homeostasis of the cardiovascular system. Failure of the interactions may lead to sympathetic hyperactivity, promoting the occurrence of life-threatening ventricular tachyarrhythmias (6), whereas augmented vagal tone may exert a protective and antiarrhythmic effect (6, 26, 31). Severe disease conditions such as acute myocardial infarction (19), panic disorder (10), and congestive heart failure (10) all exhibit signs of autonomic function imbalance. Consistent with autonomic imbalance, patients who have suffered an acute myocardial infarction have a marked decrease in heart rate (HR) variability (HRV) due to a decrease in vagal and an increase in sympathetic neural activities (14, 19, 31). The consequences of ANS dysfunction in noncardiac disease are also widely reported (3, 8, 29). A reliable way of quantifying the ANS dynamics is essential for studying ANS dysfunction-related diseases. Sympathetic nervous activities can be directly measured by assessing cardiac norepinephrine overflow to plasma by using isotope dilution (18) or via measuring muscle sympathetic nerve activity by microneurography (11). While these methods are accurate, they are invasive and preclude them from wide usage. A method, therefore, must not only be accurate to capture, quantify, and separate dynamic characteristics of the two components of the ANS, but it should be noninvasive if it is to be used as a reliable screening and diagnostic procedure of clinical importance.

Currently, the most widely used noninvasive method to assess autonomic activities is to compute the power spectral density (PSD) of HRV data. The PSD is a linear method that extracts periodic components in the frequency domain, and its popularity stems from its computational ease, provided by the fast Fourier transform (FFT). PSD of human HRV can be divided into three main frequency zones: PSD below 0.04 Hz is considered to be very low frequency (VLF), between 0.04 and 0.15 Hz are low frequency (LF), and between 0.15 and 0.4 Hz are high frequency (HF). The VLF band is believed to be related to nonneural factors, such as temperature and hormones (5). The HF band is believed to be dominated by the PNS (1, 31), whereas the LF band is believed to be mediated by both the cardiac sympathetic and parasympathetic nervous outflows. That both nervous systems moderate PSD levels in the LF band is controversial, however. Some suggest that LF activity is largely due to the SNS and that the ANS balance can be reliably calculated as the ratio of LF to HF (15, 20, 27). Current prevailing evidence, on the other hand, suggests that the vagal contributions to LF are as significant as those of the sympathetic nervous activities; consequently, the LF-to-HF ratio would be an approximation and not an accurate measure of the ANS balance (7, 12, 13). Although initial successes have been obtained in estimating sympathovagal balance by calculating a ratio between LF and HF PSD (2, 16, 19, 32), clinical use of this method has not been widespread, mainly because the LF-to-HF ratio is not a close enough approximation of the autonomic balance, for the reasons stated above. Another deficiency of the LF-to-HF ratio is that the method is linear and...
Innovative Methodology

DECOUPLING OF THE AUTONOMIC NERVOUS SYSTEM

H1476

does not properly account for nonlinear characteristics of the ANS. A plethora of recent studies have shown that the physiological mechanisms responsible for HR fluctuations have nonlinear components (31, 33).

In a previous study (34), we have detailed the technical development of the principal dynamic modes (PDM) method to address the above problem of separating the dynamics of the two nervous systems. The PDM has been shown to be a novel method for characterizing nonlinear physiological systems while reducing higher-order dimensions. The PDM is a nonlinear method because it is based on the calculation of quadratic Volterra-Wiener kernels. By using a few subjects, the feasibility of this modified PDM was demonstrated, as the first two dominant PDMs obtained from the HR data of humans corresponded well with the two autonomic nervous activities.

In this study, the aim is to fully demonstrate the reliability of the PDM, as well as to promote new research applications related to ANS dysfunction by using the PDM approach. The study is based on the participation of 28 healthy subjects who consented to administration of pharmacological blockades of the ANS. The blockades were used as a validation of the dynamics of the sympathetic and parasympathetic nervous systems obtained via the PDM during control conditions. Quantification of the ANS dynamics via the PDM is compared with the PSD to better illustrate the advantages of the PDM over the PSD.

METHODS

The experimental protocol in this study was adapted from a previous study (28).

Subjects

Twenty-eight volunteers (13 men and 15 women; mean age 30 ± 6 yr) were recruited with the aid of a Columbia Medical Center bulletin board advertisement. The candidates were examined by one of the authors (K.-M. Jan) before the study. Subjects with any active medical illness were excluded. Special attention was made to exclude subjects with a history of asthma, tachycardia, bradycardia, glaucoma, or prostate enlargement. Subjects taking any medications and smokers were also excluded.

The study protocol was approved by institutional review committees consisting of both Columbia University and State University of New York at Stony Brook. All subjects provided written informed consent for the study.

Study Protocols

All studies were performed in either the Clinical Research Center at the Presbyterian Hospital or the Intensive Care Unit Procedure Room of the Allen Pavilion at Columbia University Hospital. On the day of their study, subjects were not allowed to consume any beverages except water. Electrodes were attached to the chest surface for continuous monitoring of ECG. A sphygmomanometer cuff was attached to one arm for blood pressure monitoring. An intravenous catheter was placed in a vein of the patient’s arm aseptically for drug administrations. An ECG monitor (model 78354A, Hewlett-Packard), a blood pressure monitor (model 7000, Colin Medical Instruments), and a PowerLab data-acquisition system (PowerLab Model ML750, ADInstruments) were used to collect the ECG and radial arterial pressure signal. Throughout the study, subjects were required to breathe according to a random interval breathing tone (played by a computer, with uniform distributed random intervals of <10 s). The subjects were not allowed to talk and were told to restrain from any unnecessary movements during the experiment.

Subjects were randomly divided into two groups for experiments. Group A: supine position and parasympathetic blockade. The subjects (n = 15) were lying on a hospital bed during the experiment, and data were continuously recorded during control state and administration of autonomic blockades. After taking baseline recordings for 20 min (control state), atropine was injected (initially 0.0075 mg·kg⁻¹·min⁻¹ for 2 min followed by 0.001 mg·kg⁻¹·min⁻¹ for 23 min) to block parasympathetic nervous activity. After 5 min (to allow for physiological equilibration and to prevent transient nonstationarity in each condition), propranolol was injected (initially 2 mg·min⁻¹ for 2 min followed by 1 mg·min⁻¹ for 23 min) to block sympathetic nervous activity, resulting in complete blockade of the ANS.

Group B: upright position and sympathetic blockade. The subjects (n = 13) were standing comfortably in the upright position. Instead of atropine, propranolol was injected first followed by atropine. The duration of data collection and drug dosages were identical to those of group A.

The doses chosen for sympathetic and parasympathetic blockade with propranolol and atropine, respectively, were selected to be sufficient for complete blockade of the parasympathetic or sympathetic system (17).

Data Analysis

Measurements of ECG signals were collected with a sampling rate of 400 Hz to allow accurate detection and identification of QRS complexes in the ECG (31). The QRS complexes were used to identify beat locations. Once the timing of beats was determined, an instantaneous HR signal was created at a sampling rate of 4 Hz by using the technique described in Ref. 3. HR signals were downsampled to 1 Hz after low-pass filtering at 0.5 Hz to concentrate on the frequency bands of interest, which are all below 0.5 Hz. Furthermore, mean and trends were removed from the HR data. Segments of signals containing 600 data points, which corresponds to 10 min, were used for both the PDM and PSD analyses. Mean arterial pressure (MAP) was calculated for each of the 10-min segments.

Analyzing HR Data By Using PDM Method

The PDM method, originally introduced by Marmarelis (21), is a method based on extracting only the principal dynamic components of the signal via eigen decomposition. The PDMs are calculated using Volterra-Wiener kernels based on expansion of Laguerre polynomials (21). Among all possible choices of expansion bases, there are some that require the minimum number of basis functions to achieve a given mean-square approximation of the system output. This minimum set of basis functions is termed the PDM of the nonlinear system. This method specifically accounts for the inherent nonlinear dynamics of HR control, which the current PSD method is unable to do. A minimum set of basis functions is determined by using a method widely known as principal component analysis, in which the dominant eigenvectors and eigenvalues are retained as they relate more closely to the true characteristics of the signal, and nondominant eigenvectors and eigenvalues represent noise or nonessential characteristics. Thus principal component analysis is an approach to separate only the essential dynamic characteristics from a signal that is corrupted by noise. We have modified the PDM technique to be used with even a single output signal of HRV data, whereas the original PDM required both input and output data. A brief summary of the procedure is presented.

The accurate estimation of the Volterra-Wiener kernel requires a signal with broadband spectral characteristics. In many instances, the HR data do not exhibit broadband characteristics. Instead, significant power exists in the VLF of the HR data compared with LF and HF. Consequently, the spectral power bands of interest, the LF and HF, are dwarfed by the significantly high spectral power in the VLF band. An approach we took to reduce high spectral power in the VLF band is the method introduced by Tarvainen et al. (30), with the aim of
reducing VLF power to the level of the LF and HF bands so that overall spectral characteristics are broadband. The result of this broadening process is labeled as HRc. The PDM method requires both the input and output data, but because we have only the output signal of HR recordings, we need to create an input signal with broadband spectral characteristics. This procedure is similar to a procedure widely known as the Wiener whitening filter (23). The resulting sequence has a dynamic range similar to the HR signal; thus it is appropriate to use as an input signal. To implement this procedure, we use normalized HRc, delayed by one unit, as the input, and undelayed HRc as the output signal to estimate the Volterra-Wiener kernel. Eigen decomposition of the kernel gives eigenvalues and eigenvectors, and then a set of eigenvectors is selected according to the absolute value of respective eigenvalues to reconstruct the output signal, which is then subtracted from the HRc signal to obtain estimation error, labeled HRe. The criterion for selecting the set of eigenvectors is that they account for our set threshold value of 90% of the HR dynamics. The calculated signal, HRe, is considered to be the input signal, which has the broadband characteristics needed for accurate estimation of PDMs. The obtained input data, HRc, are then normalized to a unit variance (HRn). HRn is used as the input signal and HRc is used as the output signal to estimate the Volterra-Wiener kernel. The eigen decomposition of this kernel gives eigenvectors, which are the final values of the PDMs, and eigenvectors then represent the relative importance of each PDM. The two most dominant PDMs are selected to represent the dynamics corresponding to the sympathetic and parasympathetic nervous activities. The technical details of estimating the Volterra-Wiener kernel and extracting the PDMs from the kernel are described in the appendix.

While the obtained PDMs are in time-domain representation, we convert the PDMs to the frequency domain via the FFT to facilitate a direct comparison to the PSD. The two PDMs (Fig. 1A) have a dominant peak centered at 0.1 Hz, which is in the prescribed frequency range of the sympathetic nervous system. The second PDM (Fig. 1B) shows a prominent peak centered at 0.17 Hz as well as a secondary peak at 0.04–0.05 Hz. The significance of these two peaks in Fig. 1B is that the parasympathetic nervous system operates both in the LF and HF bands. The same observations are found with PDMs in the upright position (Fig. 1, C and D).

With application of atropine during the supine position, the PDM corresponding to the SNS (Fig. 1E) retains its magnitude in the LF range, while the PDM corresponding to the PNS (Fig. 1F) is markedly reduced compared with those of the supine control. In the upright position, with the application of propranolol, the PDM representing the SNS is significantly reduced (Fig. 1G), while the PDM representing the PNS (Fig. 1H) is negligibly affected. These phenomena are consistent with the pharmacological effects of atropine (blocks parasympathetic activities) and propranolol (blocks sympathetic activities). When both atropine and propranolol are applied (double blockade condition), the PDMs corresponding to the sympathetic [Fig. 1I (supine) and Fig. 1K (upright)] and parasympathetic [Fig. 1J (supine) and Fig. 1L (upright)] are nearly eliminated.

To facilitate a direct comparison to the PSD, the values of the PDM were scaled to represent “power” of the PDM in the

### PDM Analysis

Two dominant PDMs (with time lag $M = 60$, Laguerre coefficient $\alpha = 0.5$, and the number of Laguerre functions $L = 6$) correspond to the main frequency-response characteristics of the SNS and PNS. The criterion for selecting the above set of parameters is that a set of PDMs account for our arbitrarily set threshold value of 90% of the HR dynamics, and these chosen parameters were found to be optimal (22, 34). Furthermore, $M = 60$ was selected to ensure that all of the HR dynamics had been fully accounted for. The choice of time lag, $M = 60$, corresponds to the memory of the dynamics; thus, while a value greater than that selected will not affect the accuracy of the estimates, a value lower than that selected will result in inaccurate results. We obtained fairly consistent waveforms corresponding to the parasympathetic and sympathetic nervous activities for both groups of subjects, shown in Fig. 1, A, B, E, and F, for the supine position, and in Fig. 1, C, D, G, and H, for the upright position.

### Results

#### HR and Arterial Pressure

The mean HRs (MHRs) and MAP and their SD values during control and after application of pharmacological blockades are shown in Table 1. Compared with supine control (without drug) position, the MHR significantly increased with atropine and also with double blockade; the MHR significantly decreased from atropine to double blockade. Compared with upright control position, the MHR significantly decreased with propranolol and then increased with double blockade; the MHR also increased from propranolol to double blockade. The only statistically significant increase in the MAP was observed from supine control to application of atropine.

<table>
<thead>
<tr>
<th></th>
<th>MHR, beats/min</th>
<th>MAP, mmHg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Supine ($n = 15$)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>68.2±13.7</td>
<td>87.9±8.1</td>
</tr>
<tr>
<td>Atropine</td>
<td>103.8±16.6*</td>
<td>97.7±13.2*</td>
</tr>
<tr>
<td>Double blockade</td>
<td>97.1±14.7*</td>
<td>101.3±13.7</td>
</tr>
<tr>
<td>Upright ($n = 13$)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>80.3±9.0</td>
<td>85.4±16.7</td>
</tr>
<tr>
<td>Propranolol</td>
<td>66.9±6.4*</td>
<td>86.3±19.1</td>
</tr>
<tr>
<td>Double blockade</td>
<td>92.1±9.4*</td>
<td>88.3±17.9</td>
</tr>
</tbody>
</table>

Values are means ± SD; $n$, number of subjects. MAP, mean arterial pressure. Comparison of mean heart rate (MHR) between any 2 cases, other than double blockade to double blockade, is statistically significant (*$P < 0.001$, †$P < 0.05$).
frequency domain. This was achieved by multiplying eigenvalues associated with the PDMs followed by squaring of the results. Therefore, the area under the frequency band of interest (0.04–0.5 Hz) is considered to represent the power of the PDM. Quantification of the SNS and PNS activities from control to administration of pharmacological blockades by the power of PDM is shown in the two left columns of Table 2. Atropine significantly reduces both SNS (from a mean of 0.059 to 0.027, \( P = 0.024 \)) and PNS (from a mean of 0.483 to 0.121, \( P < 0.001 \)) compared with the control conditions. This translates into a reduction of 36% for the SNS and 71% for the PNS, with significantly greater reduction of the PNS than SNS (\( P = 0.003 \)). Propranolol also significantly reduces both the SNS (from 0.102 to 0.032, a reduction of 63%, \( P = 0.002 \)) and PNS (from 0.578 to 0.419, a reduction of 28%, \( P < 0.001 \)). However, the decrease of the SNS is significantly greater than that of the PNS (\( P < 0.001 \)).

**Power Spectral Analysis**

The averaged power spectra (solid lines) during the supine and upright positions are shown in Fig. 2, A and C, respectively. The dotted lines represent the SE bounds. Unlike the PDM, these PSD plots show composite spectral peaks associated with the parasympathetic and sympathetic nervous activities. A general trend of decreasing power is...
Table 2. Comparison of the PDM and PSD in delineating SNS and PNS activity during control and application of pharmacological blockades

<table>
<thead>
<tr>
<th>SNS (Power of PDM)</th>
<th>PNS (Power of PDM)</th>
<th>SNS-to-PNS ratio</th>
<th>LF (PSD Mean)</th>
<th>HF (PSD Mean)</th>
<th>LF-to-HF ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Supine control</td>
<td>0.059±0.013</td>
<td>0.483±0.053</td>
<td>0.113±0.018</td>
<td>47.05±12.11</td>
<td>9.19±2.19</td>
</tr>
<tr>
<td>Supine with atropine</td>
<td>0.027±0.004*</td>
<td>0.121±0.014*</td>
<td>0.280±0.092‡</td>
<td>3.29±1.21*</td>
<td>0.45±0.06*</td>
</tr>
<tr>
<td>ANS reduction by atropine, %</td>
<td>36±6</td>
<td>71±4‡</td>
<td></td>
<td>87±4</td>
<td>88±4</td>
</tr>
<tr>
<td>Upright control</td>
<td>0.102±0.024</td>
<td>0.578±0.052</td>
<td>0.157±0.025</td>
<td>86.66±19.55</td>
<td>7.42±1.63</td>
</tr>
<tr>
<td>Upright with propranolol</td>
<td>0.032±0.008*</td>
<td>0.419±0.057*</td>
<td>0.067±0.009∥</td>
<td>51.11±18.54*</td>
<td>4.13±1.12</td>
</tr>
<tr>
<td>ANS reduction by propranolol, %</td>
<td>63±6†</td>
<td>28±6</td>
<td></td>
<td>48.5±5</td>
<td>42±6</td>
</tr>
</tbody>
</table>

Values are means ± SE. ANS, autonomic nervous system. SNS and PNS are power of the principal dynamic modes (PDMs) representing sympathetic and parasympathetic nervous system contributions, respectively, in arbitrary units. LF and HF are the mean values of the power spectral density (PSD) curve in the respective low- and high-frequency ranges in units of (beats/min)². *Compared with control condition of the same body position, parameter (e.g., SNS) is significantly (P < 0.05) reduced by drugs (atropine or propranolol). ‡Percentage of SNS and PNS reduction by a drug is significantly (P = 0.003) different. ¶Change in SNS/PNS ratio (or LF/HF ratio) is significant (P < 0.001). All statistical tests are based on paired tests.

The main finding of this study is that the new nonlinear method known as the PDM is able to separate dynamics of the sympathetic and parasympathetic nervous activities. This was evidenced by the fact that the first two dominant PDMs have frequency characteristics similar to those of parasympathetic nervous system contributions, respectively, in arbitrary units. LF and HF are the mean values of the power spectral density (PSD) curve in the respective low- and high-frequency ranges in units of (beats/min)². *Compared with control condition of the same body position, parameter (e.g., SNS) is significantly (P < 0.05) reduced by drugs (atropine or propranolol). ‡Percentage of SNS and PNS reduction by a drug is significantly (P = 0.003) different. ¶Change in SNS/PNS ratio (or LF/HF ratio) is significant (P < 0.001). All statistical tests are based on paired tests.

DISCUSSION

The main finding of this study is that the new nonlinear method known as the PDM is able to separate dynamics of the sympathetic and parasympathetic nervous activities. This was evidenced by the fact that the first two dominant PDMs have frequency characteristics similar to those of parasympathetic nervous system contributions, respectively, in arbitrary units. LF and HF are the mean values of the power spectral density (PSD) curve in the respective low- and high-frequency ranges in units of (beats/min)². *Compared with control condition of the same body position, parameter (e.g., SNS) is significantly (P < 0.05) reduced by drugs (atropine or propranolol). ‡Percentage of SNS and PNS reduction by a drug is significantly (P = 0.003) different. ¶Change in SNS/PNS ratio (or LF/HF ratio) is significant (P < 0.001). All statistical tests are based on paired tests.

The main finding of this study is that the new nonlinear method known as the PDM is able to separate dynamics of the sympathetic and parasympathetic nervous activities. This was evidenced by the fact that the first two dominant PDMs have frequency characteristics similar to those of parasympathetic nervous system contributions, respectively, in arbitrary units. LF and HF are the mean values of the power spectral density (PSD) curve in the respective low- and high-frequency ranges in units of (beats/min)². *Compared with control condition of the same body position, parameter (e.g., SNS) is significantly (P < 0.05) reduced by drugs (atropine or propranolol). ‡Percentage of SNS and PNS reduction by a drug is significantly (P = 0.003) different. ¶Change in SNS/PNS ratio (or LF/HF ratio) is significant (P < 0.001). All statistical tests are based on paired tests.
and sympathetic activities. Furthermore, compared with the PSD method, the PDM provides more accurate sympathetic-to-parasympathetic ratios owing to the fact that the PDM provides better characterization of the true ANS dynamics (34). This technique allows a more in-depth analysis of the ANS and offers for the first time a more accurate measure of the balance between the two autonomic nervous activities. Furthermore, the method specifically accounts for the inherent nonlinear dynamics of HR control, which the PSD does not do.

The PDM is considered to be a general-purpose method. It was originally developed to understand dynamic characteristics of renal autoregulatory mechanisms (22). This study found that the PDM was for the first time able to separate the dynamics of the two autoregulatory mechanisms (myogenic and tubuloglomerular feedback) involved in renal autoregulation. This result was significant because the two mechanisms are known to interact in a nonlinear manner. Recently, we have modified the PDM technique to be used with even a single output signal of HRV data, whereas the original PDM required both input and output data, and the technical details involved in implementation of the method as well as a preliminary feasibility study on a limited number of subjects were provided (34). Current results based on a larger pool of subjects (n = 28) corroborate our previous finding that the first two dominant PDMs obtained from the HR data of healthy human subjects correspond to the two autonomic nervous activities. In addition, the application of the autonomic nervous blocking agents propranolol and atropine corroborated our previous finding that the magnitude of the waveforms corresponding to either sympathetic or parasympathetic activities were significantly reduced.

Furthermore, compared with propranolol, atropine should not affect the dynamics of the PDM associated with sympathetic and parasympathetic neural activities, respectively, we do observe some changes in the shape of waveforms. For example, a change in the shape of the PDM waveform associated with SNS from control to application of atropine, shown in Fig. 1, A and E, as well as a change in the shape of the PDM waveform associated with PNS from control (Fig. 1D) to application of propranolol (Fig. 1H), may reflect nonlinear interactions between the two nervous systems. The presence of interactions between the two nervous systems has been reported in many experimental studies (13, 19, 24). For example, Miyamoto et al. (24) found that β-adrenergic blockade with intravenous propranolol administration in anesthetized rabbits decreased the dynamic gain of the transfer function during vагal stimulation, which may suggest that decreased SNS may blunt the HR responses to the PNS activation. Likewise, decrease in muscle sympathetic nerve activity after atropine administration has been reported by Montano et al. (25). Another plausible explanation of atropine affecting the dynamics of the PDM associated with sympathetic nervous activities may be attributable to a significant increase in the MAP, as observed in Table 1. With increased MAP, the arterial baroreflex mechanism may operate to decrease the SNS to maintain homeostasis.

Another explanation of the change in the shape of the PDM that is not targeted by pharmacological blockade may be incomplete separation of the two nervous systems with the PDM. Note that the data analyzed contain ~10 min of ECG recordings. Given the fact that the ANS is a closed-loop system with feedback processes to maintain an optimal steady-state level, and as our data duration is long enough to complete this feedback process, incomplete separation of the ANS with the PDM is not an entirely unexpected outcome. For this scenario, the PDM is then a reflection of the dynamics pertaining to the interactions between the PNS and SNS. This may explain why, as shown in Table 2, irrespective of the pharmacological blockade, both sympathetic and parasympathetic dynamics decreased, although the percentage of reduction from the control cases was significantly greater (71% reduction of the PNS vs. 36% reduction of the SNS with atropine; 63% reduction of the SNS vs. 28% reduction of the PNS with propranolol) for the targeted nervous system. Because the PSD is not able to separate the dynamics of the two ANS mechanisms, the percentage of decrease in LF and HF power from the control conditions was similar in amount with either drug (87% reduction of the HF vs. 88% reduction of the LF with atropine; 48% reduction of the LF vs. 42% reduction of the HF with propranolol).

The results based on the PSD (see Fig. 2) lump together both sympathetic and parasympathetic nervous activities. The assumption is that LF (0.04–0.15 Hz) is predominantly sympathetic and HF (0.15–0.5 Hz) is solely mediated by the parasympathetic nervous system, although it is well established that the LF is mediated by both nervous systems (1, 9, 13). Specifically, sympathovagal balance as calculated simply by taking the LF-to-HF ratio of the PSD relies on two major inadequate assumptions: that the parasympathetic nervous system dynamics are exhibited only in high frequencies and that ANS control is linear. It has been well established that dynamics of the PNS are not only reflected in high frequencies, but they are also well represented in low frequencies (7, 9). A report corroborating this statement suggests that the LF control to application of atropine, shown in Fig. 1, the shape of the PDM waveform associated with SNS from changes in the shape of waveforms. For example, a change in the ANS and offers for the first time a more accurate measure of the balance between the two autonomic nervous activities. Furthermore, the method specifically accounts for the inherent nonlinear dynamics of HR control, which the PSD does not do.
ponent results from an interaction of both the sympathetic and parasympathetic nervous systems and not solely the sympathetic nervous activity (13). Second, the LF-to-HF ratio is based on linear power spectral analysis, which itself is limited because it is widely recognized that control of autonomic nervous systems involves nonlinear interactions. It is through efficient interactions between vagal and sympathetic nervous systems that homeostasis of the cardiovascular system is properly maintained. The interactions are believed to be nonlinear because physiological conditions would most likely involve autonomic nervous system regulation based on dynamic and simultaneous activity of the vagal and sympathetic nervous systems in response to physical environmental stress. A recent report suggests a quadratic nonlinearity between HRV and vagal effect rather than a linear relationship (9). If the LF-to-HF ratio is linear and LF and HF truly reflect sympathetic and parasympathetic nervous activities, respectively, then an autonomic blockade such as atropine should increase the LF-to-HF ratio. Clearly this is not the case, as also is evident in our PSD plot shown in Fig. 2, B and D, and Table 2; with atropine or propranolol, we obtain significant power reduction in both low and high frequencies. Therefore, it is not surprising that the LF-to-HF ratio based on the PSD, as shown in Table 2, does not truly reflect sympathovagal balance. While the PDM also shows a decrease in dynamics associated with the sympathetic and parasympathetic nervous activities with pharmacological blockades, unlike the PSD, the decrease is significantly greater for the targeted nervous system, as illustrated in Table 2.

With a sampling frequency of 1 Hz or a Nyquist frequency of 0.5 Hz, as there is no significant power in the spectrum beyond 0.4 Hz, one would expect eigenvalues corresponding to the first two PDMs to have the greatest values among the possible 61 eigenvalues \((M + 1)\). For every condition, the ratio between the sum of the two most significant PDMs and the sum of the rest of the PDMs is calculated. For most cases, the two most significant PDMs together account for \(>90\%\) of the total dynamics. Therefore, given the fact that autonomic nervous systems span most of the frequencies examined, the finding of the two most significant PDMs reflecting \(>90\%\) of the total dynamics is highly relevant.

**Response of Propranolol on the Spectral Power is Body Position Dependent**

Propranolol was administered during upright position rather than supine position. Under resting conditions, the parasympathetic tone predominates (4); thus the supine position would further accentuate dominance of the parasympathetic tone. While application of propranolol during supine position should decrease the mean HR, this does not necessarily translate into a significant decrease in the LF power. Indeed, this was observed in our study; there was no statistical difference in the area of significant decrease in the LF power. While application of propranolol during supine position should further accentuate dominance of the parasympathetic tone, predominates (4); thus the supine position would not truly reflect sympathovagal balance. While the PDM also shows a decrease in dynamics associated with the sympathetic and parasympathetic nervous activities with pharmacological blockades, unlike the PSD, the decrease is significantly greater for the targeted nervous system, as illustrated in Table 2.

**Study Limitations**

The study was based on using a random breathing technique for the purpose of obtaining a broadband spectrum of the HR signal, as this characteristic is required for computational purposes. It is our experience that in most cases normal breathing procedure provides a PSD of the HR with broadband characteristics. However, the random breathing technique ensured that the HR data would have predominantly broadband dynamic characteristics.

While in most subjects we obtained good results with atropine during supine position, we believe the unexpected decrease of the sympathetic-to-parasympathetic ratio with the PDM in 2 of the 15 subjects with atropine, as shown in Fig. 3, is primarily due to insufficient broadbandlike power spectra of the HR signal. For example, in these two subjects, with application of atropine, most of the spectral power was eliminated with only a small power concentrated at the VLF band (0 Hz to 0.04 Hz). Therefore, the broadband HR spectrum requirement of the PDM was not satisfied in these two subjects, resulting in an incorrect decrease of the sympathetic-to-parasympathetic ratio. One possible means to eliminate this source of error in future studies is to perform all experiments during upright position rather than supine position. Upright position ensures that both sympathetic and parasympathetic nervous activities are adequately represented during control condition, and subsequent stimulation or reduction of the ANS dynamics will then be more amenable to the broadband signal requirement of the PDM.

**Conclusions**

In this study, HR data from 28 normal subjects were analyzed with the recently introduced PDM to test its performance in characterizing sympathetic and parasympathetic nervous systems. The results demonstrate that the PDM is indeed able to separate dynamic characteristics of the two branches of the ANS, and the results were further validated by using pharmacological blockades of the ANS. The PSD, the current most widely used noninvasive technique in obtaining the LF-to-HF ratio for sympathovagal balance, is not able to separate the dynamics of the ANS, and consequently, far less accurate results are obtained than with the PDM. The PDM, by its nonlinear design, allows accurate separation of the dynamics of the two nervous systems and can be easily analyzed from ambulatory ECG recordings, which may have many practical applications involving autonomic dysfunction-related diseases. We have recently applied the PDM to the HRV data obtained from subjects identified with psychiatric conditions of anxiety \((n = 48)\) as well as acute emotional stress \((n = 73)\) and found that the PDM was accurate in detecting changes in the dynamics associated with the ANS (Mujica-Parodi LR, Ravindranath B, Zhong Y, Malaspina D, Klein DF, Sedler M, and Chon KH, unpublished observations). Therefore, the PDM could be potentially used as a noninvasive method for screening individuals with particular vulnerability to many dysautonomic conditions. This very possibility, of noninvasive monitoring of dysautonomia, is currently under investigation in our laboratory.
Innovative Methodology

DECOUPLING OF THE AUTONOMIC NERVOUS SYSTEM

APPENDIX

Calculation of PDMs

The estimation of PDMs using Volterra-Wiener kernels was first introduced in Refs. 21 and 22. We will briefly describe the steps involved in calculation of PDMs. For further details, the reader is referred to Refs. 21 and 22.

In discrete time, the general input-output relation of a stable (finite-memory) nonlinear time-invariant dynamic system is given by the discrete-time Volterra series:

\[
y(n) = k_0 + \sum_{m=0}^{M-1} k_1(m)x(n-m) + \sum_{m_1=0}^{M-1} \sum_{m_2=0}^{M-1} k_2(m_1, m_2)x(n-m_1)x(n-m_2) + \cdots,
\]  

where \( x(n) \) is the input and \( y(n) \) is the output of the system. \( M \) is the memory of the system. The Volterra kernels \( (k_0, k_1, k_2, \ldots) \) describe the dynamics of the system from a hierarchy of system nonlinearities.

The kernel values obtained up to a maximum lag \( M \) (kernel memory) are combined to form a real symmetric \((M+1) \times (M+1)\) square matrix:

\[
Q = \begin{bmatrix}
k_0 & \frac{1}{2} k_1(0) & \frac{1}{2} k_1(1) & \cdots & \frac{1}{2} k_1(M-1) \\
\frac{1}{2} k_1(0) & k_2(0) & k_2(0,1) & \cdots & k_2(0, M-1) \\
\frac{1}{2} k_1(1) & k_2(1,0) & k_2(1,1) & \cdots & k_2(1, M-1) \\
\vdots & \vdots & \vdots & \ddots & \vdots \\
\frac{1}{2} k_1(M-1) & k_2(M-1,0) & k_2(M-1,1) & \cdots & k_2(M-1,M-1)
\end{bmatrix}
\]

that can be used to express the second-order Volterra model response, \( \hat{y}(n) \), in a quadratic form:

\[
\hat{y}(n) = x^T(n)Qx(n),
\]

where \( T \) denotes “transpose” and the \((M+1)\)-dimensional vector \( x^T(n) = [1 \times(n) \times(n-1) \cdots \times(n-M+1)] \) is composed of the input \( M \)-point epoch at each time \( n \) and a constant 1 that allows incorporation of the lower-order kernel contributions in Eq. A.3. Expansion of the Volterra kernels on a complete basis \( [b_j(m)] \) transforms Eq. 1 into the multinomial expressions:

\[
y(n) = c_0 + \sum_{j=0}^{L-1} c_j j^v(n) + \sum_{j_1=0}^{L-1} \sum_{j_2=0}^{L-1} c_{j_1, j_2} j_1^v(n)j_2^v(n) + \cdots
\]

where

\[
v_j(n) = \sum_{m=0}^{M-1} b_j(m)x(n-m)
\]

and \( L \) is the number of Laguerre functions used. \([b_j(m)]\) are the Laguerre functions calculated with Laguerre coefficient \( x = 0.5 \). Thus \( Q \) can be constructed with the estimated kernels \( (c_0, c_1, c_2) \) in the following way:

\[
Q = \begin{bmatrix}
c_0 & \frac{1}{2} c_1 b^T \\
\frac{1}{2} Bc_1 & B^T c_1 B
\end{bmatrix}
\]

(A6)

where \( B = [b_0^T, b_1^T, \ldots, b_{(M-1)}^T] \).

Laguerre functions are chosen as an appropriate orthonormal basis because they exhibit exponential decaying properties that make them suitable for physiological systems modeling. In addition, due to basis function expansion, the estimation accuracy is maintained even with a small data length.

Because \( Q \) is a real symmetric square matrix, it can always be decomposed in the following way: \( Q = RA^T \), where the eigenvector matrix \( R \) will always be an orthonormal matrix, \( \Lambda \) is the diagonal eigenvalue matrix. We select the significant eigenvalues from \( \Lambda \) and the corresponding orthonormal eigenvectors define the PDMs of this system.

For each significant eigenvalue \( \lambda_s \), the values of the corresponding eigenvector \( \mathbf{v}_s = [\mathbf{v}_s, 0 \mathbf{v}_s, 1 \cdots \mathbf{v}_s, M] \) (with the exception of \( \mathbf{v}_s, 0 \)) define the \( s \)th PDM:

\[
p_s(m) = [\mathbf{v}_s, 1 \mathbf{v}_s, 2 \cdots \mathbf{v}_s, M]
\]

(A7)

The obtained \( s \)th PDM generates the \( s \)th mode via convolution with the input \( x(n) \). The second-order model estimation \( \hat{y}(n) \) using \( S \) PDMs is:

\[
\hat{y}(n) = \sum_{s=1}^{s} \lambda_s [p_s(m) \times x(n) + \mu_{s,0}]^2
\]

(A8)

The nonzero values \( (\mu_{s,0}) \) give rise to the contribution of linear terms.

ACKNOWLEDGMENTS

We thank Elizabeth Bobadilla for technical assistance.

GRANTS

This study was supported in part by National Heart, Lung, and Blood Institute Grant HL-76358.

REFERENCES