Simultaneous identification of static and dynamic vagosympathetic interactions in regulating heart rate

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Kawada, Toru, Masaru Sugimachi, Toshiaki Shishido, Hiroshi Miyano, Takayuki Sato, Ryochi Yoshimura, Hiroshi Miyashita, Tsutomu Nakahara, J oe Alexander, J r., and Kenji Sunagawa. Simultaneous identification of static and dynamic vagosympathetic interactions in regulating heart rate. Am. J. Physiol. 276 (Regulatory Integrative Comp. Physiol. 45): R782–R789, 1999.—We earlier reported that stimulation of either one of the sympathetic and vagal nerves augments the dynamic heart rate (HR) response to concurrent stimulation of its counterpart. We explained this phenomenon by assuming a sigmoidal static relationship between nerve activity and HR. To confirm this assumption, we stimulated the sympathetic and/or vagal nerve in anesthetized rabbits using large-amplitude Gaussian white noise and determined the static and dynamic characteristics of HR regulation by a neural network analysis. The static characteristics approximated a sigmoidal relationship between the linearly predicted and the measured HRs (response range: 212.4 ± 46.3 beats/min, minimum HR: 96.0 ± 28.4 beats/min, midpoint of operation: 196.7 ± 31.3 beats/min, maximum slope: 1.65 ± 0.51). The maximum step responses determined from the dynamic characteristics were 7.9 ± 2.9 and −14.0 ± 4.9 beats·min−1·Hz−1 for the sympathetic and the vagal system, respectively. Because of these characteristics, changes in sympathetic or vagal tone alone can alter the dynamic HR response to stimulation of the other nerve.

systems analysis; Gaussian white noise; neural network; nerve stimulation; rabbits

VARIOUS INVESTIGATORS have demonstrated complex interactions between the sympathetic and vagal systems in regulating heart rate (HR) (5, 6, 13, 14, 16, 19, 20, 23–26). Levy (13) demonstrated that concurrent sympathetic activation exaggerates the effects of vagal stimulation on the heart. This phenomenon was termed “accentuated antagonism” and was found to occur in both the static and dynamic HR responses to nerve stimulation. In the dynamic response, for example, HR was found to be greatly affected by both temporal factors (3) and the excitation sequence of the sympathetic and vagal systems (29, 30). Furthermore, we recently reported that stimulation of either the sympathetic or vagal nerve augmented the dynamic HR response to concurrent stimulation of its counterpart (9, 10). We referred to this interaction as the “bidirectional augmentation” of HR regulation and explained it by assuming a sigmoidal static relationship between autonomic nervous activity and HR.

Although nonlinear sigmoidal input-output relationships, such as the saturation phenomenon, are common in physiological systems, including blood pressure (11) and HR (15) regulations, the assumption of the sigmoidal static relationship between autonomic nervous activity and HR during dynamic stimulation of the sympathetic and vagal nerves remains to be experimentally confirmed. The term “static” in this paper indicates the absolute value of steady-state HR to a given set of tonic nerve stimulations. To our knowledge, few studies simultaneously identified the static and dynamic characteristics of HR regulation by the autonomic nervous system. The integration of the static and dynamic characteristics is essential to simulate the decoding rule of autonomic nervous activity into absolute HR value, for example, when we aim to develop a neurally regulated cardiac pacemaker (7). Thus the purpose of this study was to determine simultaneously the static and dynamic characteristics of HR regulation by the autonomic nervous system and examine to what extent the framework of a nonlinear sigmoidal relationship between autonomic nervous activity and HR can describe actual HR responses.

Although a transfer function analysis was useful in indentifying the linear input-output relationship between autonomic nervous activity and HR at a given operating point (1, 9, 10, 21), it had several limitations. First, the transfer function does not provide information about the operating point of the system, i.e., the direct current components of the input and output signals are ignored. Second, the transfer function cannot characterize the nonlinear input-output relationship of the system. These two limitations result in the third limitation: the transfer function analysis cannot combine the input-output data obtained from different operating points of the nonlinear system. Therefore, the absolute output values of the nonlinear system to given input signals cannot be predicted by the transfer function alone. To circumvent these limitations, we applied a neural network analysis to elucidate the static and dynamic characteristics of HR regulation by the autonomic nervous system. The results indicate that the nonlinear sigmoidal relationship between autonomic nervous activity and HR exists during dynamic stimulation of the sympathetic and vagal systems. Because of this nonlinear sigmoidal relationship, the
dynamic HR response to nerve stimulation crucially depends on the operating point of HR regulation.

**MATERIALS AND METHODS**

Surgical preparations. Animal care was in accordance with the guiding principles of the Physiological Society of Japan. Eight Japanese white rabbits weighing 2.3–3.3 kg were anesthetized by intravenous injection (2 ml/kg) of a mixture of urethane (250 mg/ml) and α-chloralose (40 mg/ml) and mechanically ventilated with oxygen-enriched room air. Supplemental injections (0.5 ml/kg) were given when necessary to maintain an appropriate level of anesthesia. Aortic pressure was monitored using a catheter inserted via the femoral artery. To eliminate the effects of the arterial baroreflexes, the carotid sinus and aortic depressor nerves were cut bilaterally through a midline cervical incision. The vagal nerves were also sectioned at the neck to eliminate feedback from the cardiopulmonary region. A pair of bipolar platinum electrodes were then attached to the cardiac end of the right vagal nerve for stimulation. Through a midline thoracotomy, the cardiac sympathetic nerves originating from the stellate ganglia were cut, and another pair of platinum electrodes were attached to the right cardiac sympathetic nerve for stimulation. To prevent desiccation and to provide insulation, the nerves and the stimulation electrodes were soaked in a mixture of white petrolatum (Vaseline) and paraffin. Finally, a pair of bipolar stainless steel electrodes were sutured to the right atrium to record the cardiac electrogram. The instantaneous HR was measured using a tachometer (Tachometer N4778, San-ei, Japan). During all experiments body temperature was maintained at 37°C with the use of a heating pad.

Stimulation protocols and data acquisition. The pulse duration of nerve stimulation was set at 2 ms. We adjusted the amplitude of sympathetic nerve stimulation to yield a HR increase of ~50 beats/min at a stimulation frequency of 5 Hz. After adjustment, the amplitude of sympathetic stimulation ranged from 1.8 to 3.8 V. We adjusted the amplitude of vagal nerve stimulation to yield a HR decrease of ~50 beats/min at a stimulation frequency of 5 Hz. After adjustment, the amplitude of vagal stimulation ranged from 4.2 to 6.2 V. At the end of each experiment, we confirmed that the HR response to nerve stimulation had not changed by >10%. To estimate the dynamic characteristics of HR regulation by the sympathetic and vagal systems, we used frequency-modulated, band-limited Gaussian white noise as a command signal for nerve stimulation (1, 9, 10, 17, 21). The stimulation frequency was changed every second. Hence, the input power spectrum was fairly constant up to 0.5 Hz, decreased gradually to 1/10 at ~0.8 Hz, and diminished to noise levels as the frequency approached 1 Hz.

Although in a previous study we selected relatively balanced conditions of sympathetic and vagal stimulation (10), we used submaximal stimulation frequencies in the present study to examine the entire operating range of HR regulation. Note that, however, the experiment of a single operating point with a large input amplitude is not sufficient to elucidate the characteristics of HR regulation over the entire operating range, because the low-pass filter characteristics of the system (1, 9, 10) converges the resultant HR around the middle of the operating range. Thus the study consisted of the following three protocols. In protocol 1, we stimulated the sympathetic nerve alone, using Gaussian white noise of 7±3 Hz (mean ± SD). The mean frequency was chosen to elicit maximal HR increases. In protocol 2, we stimulated the vagal nerve alone, using Gaussian white noise of 15±7 Hz. The mean frequency was set as high as possible without engendering second-degree atrioventricular block. In protocol 3, we stimulated both the sympathetic and vagal nerves, using separate independent Gaussian white noise signals (7±3 Hz for the sympathetic stimulation and 15±7 Hz for the vagal stimulation). After reaching steady state in each of the stimulation protocols, we recorded the HR response to sympathetic and vagal stimulation. In this study, the end of each experiment, we confirmed that the HR approach. We also changed the Gaussian white noise signals used among the animals. The stimulation command signals and the HR for 10 min. We randomized the order of stimulation protocols among the animals to reduce the likelihood of bias or systematic error in our identification approach. Data analysis. To analyze the data combined from the three protocols, we adopted the framework of a simple two-layered neural network. As shown in Fig. 1, the first layer of the neural network received the time series of command signals for sympathetic stimulation [s(n), s(n−1),..., s(n−k)] and vagal stimulation [v(n), v(n−1),..., v(n−k)]. Each of the time series was multiplied by the weight matrices relating to either the sympathetic system [wS(0), wS(1),..., wS(k)] or the vagal system [wV(0), wV(1),..., wV(k)]. In this structure, the weight matrices correspond to the impulse responses of the sympathetic and vagal systems, respectively. We then performed mathematical transformation on the output from the first layer according to a function defined in the second layer. Thus the overall mathematical description of the neural network is as follows

\[
HR_p(n) = f \left( \sum_{i=0}^{k} [w_S(i) \times s(n-i) + w_V(i) \times (n-i)] \right)
\]

where \(HR_p(n)\) is the output of the neural network, i.e., the predicted HR, and \(f(x)\) is the function defined in the second layer. \(n\) and \(k\) indicate a current data point and the length of impulse response of the system, respectively. Initially, we used a linear function in the second layer

\[
f(x) = ax + b
\]

in which no mathematical assumptions were made about the nonlinearity of the system. This function was used to elucidate the nature of nonlinearity in the system by plotting
scattergrams of the measured versus the linearly predicted HR responses. After the scattergrams revealed sigmoidal nonlinearity, we used a sigmoidal function for the second layer and estimated its defining parameters. The sigmoidal function is described as follows (11)

\[
f(x) = \frac{P_1}{1 + \exp[P_2 \times (P_3 - x)]} + P_4
\]

where \(P_1\) is the response range (the difference between the maximum and minimum HRs), \(P_2\) is the coefficient of slope, \(P_3\) is the midpoint of operation (the midpoint of the entire operating range), and \(P_4\) is the minimum HR. The midpoint of HR response is calculated as \((P_2/2 + P_4)\). The maximum slope (slope at the steepest point of the sigmoidal function) is calculated as \((P_1 \times P_2/4)\).

We provided 30 s of input data to the neural network to yield one output value, i.e., the expected impulse response of the system was at most 30 s. This length was determined on the basis of the transfer function of the system estimated in our previous studies (9, 10). We used data resampled at a rate of 2 Hz to reduce the burden of computation. This resampling did not deteriorate the estimation of the system characteristics, because both the sympathetic and vagal systems lose their gain >1 Hz in controlling HR in rabbits (9, 10, 21). To estimate the 30-s impulse response, we trained the neural network using 12 min of input-output data compiled from the first half of the three protocols in each animal. We used MATLAB (Math Works, Natick, MA) neural network toolbox for the neural network calculations. The neural network toolbox only provides a “logsig” neuron where the output of the sigmoidal neuron is limited in the range between 0 and 1. To match the output of the neural network to the measured HR, we added a “purelin” neuron to linearly scale the output from the logsig neuron. In this sense, the neural network used was “three layered” rather than “two layered.”

Statistics. After training of the neural network yielded convergence on a consistent set of weights (usually within 20 epochs), validation was made by predicting the HR response using 12 min of input data compiled from the second half of the three protocols. To quantitatively measure the accuracy of estimation, we calculated the standard error of the estimate (SEE) using the following equation

\[
\text{SEE} = \sqrt{\frac{\sum_{n=1}^{N} [\text{HR}_{P}(n) - \text{HR}(n)]^2}{N}}
\]

where \(\text{HR}_P(n)\) is the output of the neural network, and \(\text{HR}(n)\) is the measured HR. \(N\) represents the total number of output data points. The difference between the SEEs derived from the linear neural network and the nonlinear neural network was tested for significance using the Wilcoxon signed-rank test (4) for both the training and validation stages. The difference was considered to be significant for \(P < 0.05\) (\(P = 0.024\)).

The difference between the midpoint of operation and the midpoint of HR response determined by the sigmoidal function used in the second layer of the nonlinear neural network was tested using a paired t-test (4) with a significance level at \(P < 0.05\).

RESULTS

Figure 2 shows typical recordings of sympathetic stimulation (Fig. 2, top), vagal stimulation (Fig. 2, middle), and HR response (Fig. 2, bottom). Each panel comprises data obtained from protocol 1 (Fig. 2, left), protocol 2 (Fig. 2, middle), and protocol 3 (Fig. 2, right). When we stimulated the sympathetic nerve alone, the mean level of HR increased to 289 beats/min. Because HR reached a saturation zone, the dynamic HR response to sympathetic stimulation was small. When we stimulated the vagal nerve alone, the mean level of HR decreased to 110 beats/min. Although HR changed dynamically in response to vagal stimulation, lower HR seemed to be truncated at ~80 beats/min, suggesting a nonlinearity of the vagal effect on HR. When we stimulated both the sympathetic and vagal nerves, the mean level of measured HR was 167 beats/min. In this protocol, the dynamic HR response was the largest of the three protocols.

Figure 3 shows typical results of the measured HR (Fig. 3, top), the HR predicted by the linear neural

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Fig. 2. Representative recording of sympathetic (top) and vagal (middle) stimulation signals based on Gaussian white noise and HR response (bottom). Recording is comprised of segments from the sympathetic stimulation protocol (left), the vagal stimulation protocol (middle), and the simultaneous sympathetic and vagal stimulation protocol (right), bpm, Beats/min.
network (Fig. 3, middle), and the HR predicted by the nonlinear neural network (Fig. 3, bottom) obtained during the training stage. In protocol 1 (Fig. 3, left), the linear estimation showed greater variation than the measured HR (Fig. 3, insets, shows enlarged ordinate). The amplitude of the dynamic HR response of nonlinear estimation was comparable to the measured one. In protocol 2 (Fig. 3, middle), the linear estimation did not reproduce the truncation phenomenon observed in the measured HR response, while the nonlinear estimation did. In protocol 3 (Fig. 3, right), both the linear and nonlinear estimations showed HR response comparable to the measured one. If we examined precisely, however, the amplitude of linear estimation was smaller than that of measured HR. The nonlinear estimation reproduced the measured HR response reasonably well.

Figure 4 shows the scattergrams derived from the representative data presented in Fig. 3. Figure 4, left and right, depicts the measured HRs versus the predicted HRs determined by the linear and nonlinear neural networks, respectively. Figure 4, left, clearly indicates the existence of a nonlinear sigmoidal relationship between the linearly predicted and the measured HRs. As expected, when we used a nonlinear sigmoidal function for the second layer, the relationship between the predicted and the measured HRs became fairly linear. The SEE of nonlinear estimation was significantly smaller than that of linear estimation during both the training stage (9.1 ± 1.9 vs. 13.7 ± 3.4 beats/min, \( P < 0.05 \)) and the validation stage (11.3 ± 2.9 vs. 15.4 ± 3.9 beats/min, \( P < 0.05 \)). The parameters of the estimated sigmoidal function of the nonlinear neural network are provided in Table 1. The midpoint of HR response was significantly lower than the midpoint of operation (\( P < 0.05 \)).

Figure 5 shows the step responses of HR to sympathetic stimulation (Fig. 5A) and vagal stimulation (Fig. 5B) averaged over all the animals. The step responses were calculated by integrating the respective weight matrices of the nonlinear neural network. A stepwise increase in sympathetic stimulation frequency by 1 Hz resulted in a HR increase with a maximum response of 7.9 ± 2.9 (mean ± SD) beats/min and a 90% rise time of 12.6 ± 4.9 s. A stepwise increase in vagal stimulation frequency by 1 Hz caused a decrease in HR with a maximum (negative) response of -14.0 ± 4.9 beats/min and a 90% rise time of 3.2 ± 1.2 s. The 90% rise time is defined as the time at which 90% of the maximum step response was reached. Note that the magnitudes of the step responses may be modified by the nonlinear sigmoidal function in the second layer and are thus dependent on the particular operating point of the HR regulation.
We have determined the static and dynamic characteristics of HR regulation by the autonomic nervous system during dynamic stimulation of the sympathetic and vagal systems. Because of a sigmoidal static relationship between nerve stimulation and HR, under relatively balanced conditions of sympathetic and vagal system tones, simultaneous stimulation of the sympathetic and vagal nerves augmented the dynamic HR response in comparison to what would be observed by stimulation of either nerve alone.

Bidirectional augmentation through a nonlinear sigmoidal input-output relationship. In a previous paper, we demonstrated that the dynamic HR response to either sympathetic or vagal nerve stimulation was augmented by simultaneous tonic stimulation of the other nerve (9). The augmentation of HR response also occurred when the other nerve was dynamically stimulated, irrespective of stimulation patterns (10). We referred to this phenomenon as the bidirectional augmentation of HR regulation and explained it by assuming a sigmoidal static relationship between autonomic nervous activity and HR. To summarize our interpretations briefly, whenever we stimulated the sympathetic nerve or the vagal nerve alone, mean HR deviated from the steepest region of the sigmoidal function, resulting in a loss of gain in HR regulation. Simultaneous stimulation of the sympathetic and vagal nerves moved the operating point back to the steepest region of the sigmoidal function, thereby augmenting the dynamic HR response to nerve stimulation.

The scattergram of the measured versus the linearly predicted HRs (Fig. 4, left) revealed the sigmoidal nonlinearity in HR regulation. When we included the sigmoidal nonlinearity in the prediction of HR, the scattergram of the measured versus the predicted HRs became fairly linear (Fig. 4, right). In accordance with the estimated parameters of the sigmoidal function, the midpoint of HR response was significantly lower than the midpoint of operation (Table 1). In other words, in terms of mean level of HR, HR responses to combined sympathetic and vagal stimulation were substantially more negative than the algebraic sum of individual responses. This phenomenon is known as the accentuated antagonism (3, 13, 25).

Interactions between the sympathetic and vagal systems in HR regulation. The sympathetic and vagal systems are known to interact in HR regulation through both prejunctional and postjunctional mechanisms. In prejunctional interaction, acetylcholine released by vagal stimulation inhibits the release of norepinephrine by acting on muscarinic receptors on the sympathetic nerve terminals (16, 20). On the other hand, sympathetic stimulation inhibits the release of acetylcholine by acting on α₁-adrenergic receptors on the vagal terminals (18, 28), although guinea pigs lack this mechanism (16). Neuropeptide Y released from sympathetic nerve terminals also interacts with acetylcholine; the former inhibits the vagal effect on the heart through mechanisms other than α₁- and β-adrenergic receptors (19, 22, 23, 26). However, it was also reported that simultaneous vagal stimulation prevented the release of neuropeptide Y from sympathetic nerve terminals (24, 29). At present, at least five distinct neuropeptide Y receptors have been identified by molecular cloning technique (2). A postjunctional interaction was evidenced by the fact that the effects of acetylcholine were predominant over those of norepinephrine on the sinoatrial pacemaker frequency in isolated rat atria (6). In the postjunctional interaction, acetylcholine inhibited the effects of sympathetic agents by attenuating the rise in intracellular concentrations of cAMP (27). This antagonism may also be partly mediated by a rise in intracellular cGMP (27).

Because these studies have indicated that interactions between the sympathetic and vagal systems are mutually inhibitory, the phenomenon of bidirectional augmentation of HR regulation could not be directly explained. However, a shift in the operating point of HR regulation through the mutually inhibitory interactions in conjunction with the nonlinear sigmoidal relationship between autonomic nervous activity and HR,

Table 1. Parameters of the sigmoidal function determined for the second layer of the nonlinear neural network

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Response range, beats/min</td>
<td>212.4 ± 46.3</td>
</tr>
<tr>
<td>Coefficient of slope</td>
<td>0.035 ± 0.021</td>
</tr>
<tr>
<td>Minimum HR, beats/min</td>
<td>96.0 ± 28.4</td>
</tr>
<tr>
<td>Midpoint of operation, beats/min</td>
<td>196.7 ± 31.3</td>
</tr>
<tr>
<td>Maximum slope</td>
<td>1.65 ± 0.51</td>
</tr>
</tbody>
</table>

Data are means ± SD. HR, heart rate.

DISCUSSION

We have determined the static and dynamic characteristics of HR regulation by the autonomic nervous system during dynamic stimulation of the sympathetic and vagal systems. Because of a sigmoidal static relationship between nerve stimulation and HR, under relatively balanced conditions of sympathetic and vagal system tones, simultaneous stimulation of the sympathetic and vagal nerves augmented the dynamic HR response in comparison to what would be observed by stimulation of either nerve alone.

Bidirectional augmentation through a nonlinear sigmoidal input-output relationship. In a previous paper, we demonstrated that the dynamic HR response to either sympathetic or vagal nerve stimulation was augmented by simultaneous tonic stimulation of the other nerve (9). The augmentation of HR response also occurred when the other nerve was dynamically stimulated, irrespective of stimulation patterns (10). We referred to this phenomenon as the bidirectional augmentation of HR regulation and explained it by assuming a sigmoidal static relationship between autonomic nervous activity and HR. To summarize our interpretations briefly, whenever we stimulated the sympathetic nerve or the vagal nerve alone, mean HR deviated from the steepest region of the sigmoidal function, resulting in a loss of gain in HR regulation. Simultaneous stimulation of the sympathetic and vagal nerves moved the operating point back to the steepest region of the sigmoidal function, thereby augmenting the dynamic HR response to nerve stimulation.

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might cause the synergistic interactions between the sympathetic and vagal systems.

Simulation of the static HR response using identified system characteristics. To explore the outcome of the nonlinear sigmoidal relationship between autonomic nervous activity and HR, we simulated the static HR response to a variety of combinations of tonic sympathetic stimulation (0, 2, 4, 6, and 8 Hz) and tonic vagal stimulation (0, 5, 10, 15, and 20 Hz). In each of the combinations, we generated 30 s of stimulation command signals and calculated the HR response by using the trained nonlinear neural network. The HR at the end of the 30 s was used as the static HR response.

Illustrated in Fig. 6 is an averaged response surface of HR to various combinations of tonic sympathetic and vagal stimulations. When there is no sympathetic stimulation, HR decreases along the vagal axis (Fig. 6, curve a-b). The decreasing slope becomes steeper as the frequency of concomitant sympathetic stimulation increases (Fig. 6, curve c-d). When there is no vagal stimulation, HR increases along the sympathetic axis (Fig. 6, curve a-c). The increasing slope becomes shallower as the frequency of concomitant vagal stimulation increases (Fig. 6, curve b-d). These general characteristics are consistent with the phenomenon of accentuated antagonism. The similar response surface was reported in experiments in dogs using actual tonic stimulation of the sympathetic and vagal nerves (15).

Besides these characteristics consistent with the accentuated antagonism, the response surface revealed a particular region (the region circumscribed by dashed line in Fig. 6) where the slope of HR response to sympathetic stimulation becomes steeper with concomitant vagal stimulation (Fig. 6, line 3–4 vs. 1–2). In this region, the slope of HR response to vagal stimulation also becomes steeper with concomitant sympathetic stimulation (Fig. 6, line γ–δ vs. α–β). The HR response falls to near its baseline level (HR in the absence of any nerve stimulation) despite simultaneous stimulation of both nerves. This simulation result reinforces our previous conclusion that bidirectional augmentation of HR regulation occurs under relatively balanced conditions of sympathetic and vagal stimulation (9, 10). Therefore, the relatively simple two-layered structure with a sigmoidal function was able to integrate the concepts of both bidirectional augmentation and accentuated antagonism.

Neural network analysis. In this study, we made use of a neural network, together with Gaussian white noise, to identify the characteristics of the static and dynamic HR response to sympathetic and vagal stimulation over the entire operating range. Step responses of HR derived from the weight matrices of the nonlinear neural network (Fig. 5) were consistent with those estimated by transfer function analysis in our previous studies (9, 10). Thus the neural network was effective in analyzing not only the static nonlinear characteristics but the dynamic linear transfer characteristics of the system as well. Because the stimulation command signal of Gaussian white noise was rich in frequency components, the neural network received sufficient information regarding input-output relationships to be able to provide an unbiased estimation of system characteristics.

As mentioned in MATERIALS AND METHODS, we had to analyze combined data obtained from different operating points to elucidate the characteristics of HR regulation over the entire operating range. Although the transfer function analysis may be applicable to elucidate the linear input-output relationship of the system in each protocol, it appears to be difficult to reconstruct the whole system characteristics from the estimated transfer functions. In contrast to the transfer function analysis, because the neural network analysis can carry information about the operating point of the system, we can successfully estimate the whole system characteristics over the entire operating range.

Our application of the nonlinear neural network algorithm enabled greater precision than performing sequential estimations of the weight matrices and the sigmoidal function by least-square fitting. Because the nonlinear neural network could estimate the weight matrices of the first layer and the sigmoidal function of the second layer simultaneously, an improved estimation accuracy for one layer necessarily means an improved estimation accuracy for the other.

Limitations. Although the nonlinear neural network had a lower SEE than the linear neural network in the prediction of HR response, there were still substantial differences between the predicted and measured HRs. Although using other nonlinear functions (an asymmetric sigmoidal function, etc.) might further reduce the SEE, the symmetric sigmoidal function was most relevant for physiological interpretation of the identified parameters (11). Increasing the number of neurons...
might also reduce the SEE. Although we have tried the nonlinear neural network with two neurons in the second layer, it failed to reduce the SEE in the validation stage.

We investigated the HR response to nerve stimulation in anesthetized rabbits. Because we cut both the sympathetic and vagal nerves, the effects of anesthesia on the central nervous system would affect the present results only minimally. However, the operating point of HR regulation under conscious conditions depends on tonic activity of the sympathetic and vagal systems that are controlled by the central nervous system. Further studies are needed to understand autonomic control of HR under conscious conditions.

Finally, the stimulating pattern of Gaussian white noise is different from the physiological discharge of autonomic nerves (12). Notably, the aphasic nature of Gaussian white noise stimulation relative to each R-R interval would mask a phase-dependent sensitivity of HR response to nerve stimulation (8). Moreover, we stimulated the whole bundle of efferent nerve fibers together. Thus the HR response in this study does not account for possible regional differences in nerve function among the nerve fibers.

In conclusion, this study demonstrated the existence of a nonlinear sigmoidal relationship between autonomic nervous activity and HR during dynamic stimulation of the sympathetic and vagal systems. Because of this sigmoidal nonlinearity, changes in sympathetic or vagal system tone alone could alter the dynamic HR response to stimulation of the other nerve. Under balanced conditions of sympathetic and vagal system tone, this nonlinearity results in bidirectional augmentation of HR regulation.

Perspectives

We previously reported the possibility of developing a neurally regulated cardiac pacemaker that translated the sympathetic nervous activity into HR by using a simple two-layered neural network. The prediction error of HR response during validation stage (11.3 ± 2.9 beats/min) was only 5 and 7% of the response range and midpoint of response of HR regulation, respectively. This range of prediction error might be within the acceptable range for physiological regulation of HR by the autonomic nervous system. Because the weight matrices in the first layer corresponded to impulse responses of the sympathetic and vagal systems in regulating HR, respectively, the neural network had a relatively simple structure comprising two linear transfer functions followed by a nonlinear sigmoidal function. Therefore, it is conceivable that we can implement the decoding rule from autonomic nervous activity to HR into the artificial cardiac pacemaker.

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