Investigating Feed-Forward Neural Regulation of Circulation From Analysis of Spontaneous Arterial Pressure and Heart Rate Fluctuations

Jacopo M. Legramante, MD; Gianfranco Raimondi, MD; Michele Massaro, MD; Salvatore Cassarino, MD; Giuseppe Peruzzi, MD; Ferdinando Iellamo, MD

Background—Analysis of spontaneous fluctuations in systolic arterial pressure (SAP) and pulse interval (PI) reveals the occurrence of sequences of consecutive beats characterized by SAP and PI changing in the same (+PI/+SAP and −PI−SAP) or opposite (−PI/+SAP and +PI−SAP) direction. Although the former reflects baroreflex regulatory mechanisms, the physiological meaning of −PI/+SAP and +PI−SAP is unclear. We tested the hypothesis that −PI/+SAP and +PI−SAP “nonbaroreflex” sequences represent a phenomenon modulated by the autonomic nervous system reflecting a feed-forward mechanism of cardiovascular regulation.

Methods and Results—We studied anesthetized rabbits before and after (1) complete autonomic blockade (guanethidine+propranolol+atropine, n = 13; CAB), (2) sympathetic blockade (guanethidine+propranolol, n = 15; SB), (3) parasympathetic blockade (atropine, n = 16), (4) sinoaortic denervation (n = 10; SAD), and (5) controlled respiration (n = 10; CR). Nonbaroreflex sequences were defined as ≥ 3 beats in which SAP and PI of the following beat changed in the opposite direction. CAB reduced the number of nonbaroreflex sequences (19.1 ± 12.3 versus 88.7 ± 36.6, P < 0.05), as did SB (25.3 ± 11.7 versus 84.6 ± 23.9, P < 0.001) and atropine (11.2 ± 6.8 versus 94.1 ± 32.4, P < 0.05). SB concomitantly increased baroreflex sensitivity (1.18 ± 0.11 versus 0.47 ± 0.09 ms/mm Hg, P < 0.01). SAD and CR did not significantly affect their occurrence.

Conclusions—These results suggest that nonbaroreflex sequences represent the expression of an integrated, neurally mediated, feed-forward type of short-term cardiovascular regulation able to interact dynamically with the feedback mechanisms of baroreflex origin in the control of heart period. (Circulation. 1999;99:1760-1766.)

Key Words: nervous system, autonomic ■ baroreceptors

In the intact circulation, beat-by-beat spontaneous fluctuations in RR interval have been shown to be linked to beat-by-beat changes in arterial pressure through baroreflex mechanisms. Analysis of the continuous relationship between systolic arterial pressure (SAP) and RR interval revealed that spontaneous increases or decreases in SAP induce directionally similar reflex changes in RR interval.1 On this basis, a new technique, called spontaneous baroreflex, has been developed for dynamically studying the arterial baroreflex control of the sinus node.2 This method, used in a number of laboratories,3,4 is based on the computer scanning of SAP and RR interval (or pulse interval, PI) time series to identify sequences of spontaneously occurring consecutive beats in which SAP and PI of the following beat change in the same direction, ie, hypertensive/brady- and hypotensive/tachycardic sequences.

Beat-by-beat analysis of the continuous relationship between spontaneous fluctuations in SAP and RR interval also reveals the occurrence of sequences of consecutive beats in which SAP and RR interval (or PI) of the following beat change in the opposite direction (ie, hypertensive/tachycardic and hypotensive/brady-sequences). These sequences have been defined as nonbaroreflex, but their physiological meaning and thus their possible role in the cardiovascular regulation have not been established.

Cardiovascular neural regulation involves both central controllers and peripheral reflex mechanisms operating with feedback features (eg, arterial baroreflexes) that interact in modulating the dynamic heart rate (HR) and arterial pressure fluctuations. However, studies performed in both anesthetized and conscious animals5,6 have indicated that reflex mechanisms operating with positive feedback features could contribute to the neural regulation of the cardiovascular system and to the modulation of HR and arterial pressure variability.7 Moreover, feed-forward mechanisms of cardiovascular regulation operating through mechanically coupled changes in systolic arterial pressure and HR have also been described in humans,8–10.
On the basis of the above considerations, the concomitant occurrence of nonbaroreflex with baroreflex sequences suggests the intriguing possibility that nonbaroreflex sequences might represent an expression of autonomic regulatory mechanisms operating with feed-forward (or positive feedback) features as opposed to negative feedback mechanisms, mediated mainly by the arterial baroreflexes. Accordingly, the aim of this study was to test the hypothesis that the spontaneously occurring nonbaroreflex sequences represent a physiological phenomenon modulated by the autonomic nervous system, reflecting a feed-forward mechanism of cardiovascular regulation.

Methods

General Procedures

The study was performed on 64 adult New Zealand rabbits of both sexes (weight, 2.5 to 3.5 kg). After induction of anesthesia by ketamine (30 mg/kg IM), the ear marginal vein was cannulated for drug infusion. Afterward, the rabbits were anesthetized with α-chloralose (25 mg/kg) and urethane (250 mg/kg IV). Tracheotomy was then performed, and the trachea was intubated with a polyethylene cannula of the same diameter for measurement of respiratory flow. Rectal temperature was monitored continuously by a thermocouple (Hewlett Packard 21058 A) and maintained at 38 ±0.5°C by means of a steel hot plate on the operating table and infrared lamps. Heparin was given (10 mg/kg IV, with supplemental hourly doses of 2 mg/kg) to prevent blood clotting.

Sinoaortic Denervation

The vagus nerves and carotid bifurcations were exposed through a midline incision in the neck. The infraparotid cervical vagi were intubated within the common (2% solution)/bicarbonate (10-mEq/mL) solution to prevent blood clotting. Blood pressure transducer (P23XL). HR was monitored by a tachograph triggered by the pressure pulse or by an ECG (Gould 13-G4615-65A).

Recorded Variables

Arterial pressure and PI were continuously recorded from the femoral artery via a polyethylene cannula connected to a Gould pressure transducer (P23XL). HR was monitored by a tachograph triggered by the pressure pulse or by an ECG (Gould 13-G4615-65A).

The respiratory airflow was recorded with a pneumotachograph (Gould Fleish 000-369500) connected to the tracheal cannula and to a differential gas pressure transducer (Validyne DP45) that was integrated with a Gould integrator amplifier (13-G4615-70) to give tidal volume (VT) and pulmonary ventilation (Ve). Both inspiratory and expiratory waveforms of the respiratory cycle were integrated and recorded. Breathing frequency (f) was calculated from the interbreath period.

Arterial blood O2 and CO2 partial pressure (Pao2 and Paco2) and arterial pH (pHr) were determined at 37°C by use of a blood gas analyzer (AVL 330); the results were corrected to the animal temperature. PaO2, PaCO2, and pHr were measured at the beginning of the experiment and maintained within the following ranges: PaO2, ∼90 mm Hg; PaCO2, 28 to 36 mm Hg; and pHr, 7.35 to 7.40. When necessary, PaO2 was increased by enriching the inspired air with O2 supply, and pH was corrected by infusing a 10-mEq/mL solution of sodium bicarbonate.

Sequences Analysis

The beat-by-beat time series of SAP and PI were analyzed by a computer to identify spontaneously occurring sequences of ≥3 consecutive beats in which SAP and PI of the following (ie, lag 1) beat changed in the opposite direction, eg, SAP increasing and PI decreasing (ie, hypertension and tachycardia). These sequences were identified as nonbaroreflex sequences (Figure 1). Simultaneously, the sequences characterized by ≥3 consecutive beats in which SAP and PI of the following beat spontaneously changed in the same direction, either increasing or decreasing (ie, baroreflex sequences), were also searched (Figure 1). The threshold change between 2 consecutive SAP or PI values was set at 0.3 mm Hg and 0.6 ms, respectively. Linear regression was applied to each individual sequence, similar to the Oxford technique that uses bolus injections of vasoactive drugs. Only those sequences in which r2 >0.85 were accepted to minimize the possibility of counting a sequence in which random variations in SAP and PI appeared as a sequence. The number of nonbaroreflex sequences was calculated. Separate calculations were made for sequences in which SAP increased and PI decreased (−PI(+SAP)) and for those in which SAP decreased and PI increased (+PI(−SAP)).

The beat-by-beat time series of SAP and PI were also searched for sequences ≥3 consecutive beats in which SAP and PI of the same beat (ie, lag 0) changed in the opposite direction. The number of these sequences also was calculated.

A separate analysis of baroreflex sequences in which SAP and PI increased (+PI(+SAP)) and decreased (−PI(−SAP)) was also performed. The mean individual slope of the baroreflex sequences, obtained by averaging all slopes computed within a given experimental period, was calculated and taken as a measure of the integrated spontaneous baroreflex sensitivity and the gain in nonbaroreflex mechanisms, respectively, for that period.

Experimental Protocol

Experiments were performed on 5 separate groups of rabbits. After any surgical procedure, the rabbits were allowed to stabilize for 30 minutes before experiments began.

Group 1

Thirteen rabbits were studied under baseline conditions and after complete autonomic pharmacological blockade (guanethidine 2.5 mg/kg IV plus propranolol 2 mg/kg IV and atropine 2.5 mg/kg IV). The effectiveness of complete autonomic blockade was tested by measuring the cardiovascular responses to ephedrine (5 mg/kg IV), isoproterenol (8 μg/kg IV), and acetylcholine (1 to 2 μg/kg IV), and only the rabbits in which the responses were abolished by the blocking drugs were accepted for the study.

Group 2

Fifteen rabbits were studied under baseline conditions and after parasympathetic pharmacological blockade (guanethidine 2.5 mg/kg IV plus propranolol 2 mg/kg IV). The effectiveness of parasympathetic blockade was tested by measuring the cardiovascular responses to ephedrine (5 mg/kg IV) and isoproterenol (8 μg/kg IV), and only the rabbits in which the responses were abolished by the blocking drugs were accepted for the study.

Group 3

Sixteen rabbits were studied before and after sympathetic pharmacological blockade (atropine 2.5 mg/kg IV). The effectiveness of sympathetic blockade was tested by measuring the cardiovascular responses to ephedrine (5 mg/kg IV) and isoproterenol (8 μg/kg IV), and only the rabbits in which the responses were abolished by the blocking drugs were accepted for the study.

Group 4

Ten rabbits were studied before and after sinoaortic denervation (SAD). The effectiveness of SAD was indicated by abolition of the bradycardic response to intravenous bolus injections of phenylephrine after the surgical procedure compared with control (−77.6 ±8.5 versus −7.4 ±3.2 bpm).

Group 5

To evaluate the possible influence of respiration on the expression of the nonbaroreflex sequences, 10 rabbits were studied under control conditions and during controlled respiration, obtained by artificially ventilating the animals by means of an automatic ventilator (Small Animal Ventilator, Harvard) after they had been paralyzed with...
triethiodide gallamine (3 to 5 mg/kg IV). The animals were studied under an artificially maintained V E similar to that recorded while the animals were breathing spontaneously and under V E levels artificially increased up to 25% of the values observed in each animal during spontaneous breathing by increasing either f or VT separately.

**Data and Statistical Analyses**

All circulatory and respiratory variables were recorded on a Hewlett Packard 8-channel magnetic tape recorder (3968A) and on a Gould 8-channel polygraph (TA 4000). The data were stored and analyzed by a computerized online system for biological data elaboration. The latter has a hardware signal-conditioning system set up in our laboratory that detects the RR and/or PI by means of a dedicated trigger (set on the ECG signal and/or the arterial pressure wave, respectively) and can nominally discriminate minute differences between systolic pressure peaks, allowing a maximal error of 25 µs (equivalent to a sample frequency of 40 kHz). A second parallel hardware conditioning block detected the systolic peak by means of a peak detector that allows a maximal error of 0.06 mm Hg and is independent of sample frequency, depending only on the resolution of the AD converter (in our case, 12 bit).

Reported data were calculated from SAP and PI time series length of 4500 beats in each experimental period. The significance of the differences in the reported data between control and test periods within each experimental groups was evaluated by Student’s t test for paired observations. One-way ANOVA was used to compare changes in the number of nonbaroreflex sequences occurring after sympathetic versus parasympathetic pharmacological blockade experiments. A paired t test was also used to evaluate the significance of differences in the number of lag 1 versus lag 0 sequences. All data are presented as mean±SEM. A value of \( P<0.05 \) was considered statistically significant.

**Results**

Complete autonomic blockade induced a significant decrease in arterial pressure and HR (Table 1). Under control conditions, before complete autonomic blockade, the number of spontaneous nonbaroreflex sequences did not differ significantly on going from lag 1 to lag 0 (88.7±36.6 versus 133.4±48.2, \( P=0.27 \)). The number of nonbaroreflex sequences (lag 1) was significantly decreased by complete autonomic blockade (Figure 2). This reduction was due to a significant and marked decrease in the number of both the hypertensive/tachycardic (\( +\text{PI}/-\text{SAP} \)) and the hypotensive/bradycardic (\( -\text{PI}/+\text{SAP} \)) sequences (Table 2). As shown in Figure 2, the mean slope of the nonbaroreflex sequences did not show significant changes. Similarly, also the number of lag 0 sequences was significantly decreased by complete autonomic blockade (Figure 3).

Selective sympathetic blockade induced a significant decrease in the number of nonbaroreflex sequences (Figure 2), which involved both the \( -\text{PI}/+\text{SAP} \) and \( +\text{PI}/-\text{SAP} \) sequences (Table 2).

The mean slope of the baroreflex sequences showed a significant increase after sympathetic blockade, rising from 0.47±0.09 to 1.18±0.11 ms/mm Hg (\( P<0.01 \)), indicating an increased gain (or sensitivity) in the baroreflex mechanisms controlling heart period. The increase in baroreceptor–cardiac reflex sensitivity was significant in response to both SAP increases and decreases (ie, activating and deactivating stimuli). The mean slope of the \( +\text{PI}/+\text{SAP} \) and \( -\text{PI}/-\text{SAP} \) sequences increased from 0.51±0.10 to 1.36±0.18 ms/mm Hg (\( P<0.01 \)) and from 0.40±0.06 to 1.03±0.15 ms/mm Hg, (\( P<0.01 \)), respectively. On the contrary, the mean slope of the nonbaroreflex sequences did not show significant changes (Figure 2).
After parasympathetic blockade, HR significantly increased, whereas arterial pressure slightly decreased (Table 1). After atropine, the number of the nonbaroreflex sequences was significantly reduced (Figure 2), and again this reduction involved both the $-\Pi/1 + SAP$ and $+\Pi/2 - SAP$ sequences (Table 2). The mean slope of the nonbaroreflex sequences did not show significant changes (Figure 2).

The mean change in the number of nonbaroreflex sequences in response to the sympathetic blockade ($-59.2 \pm 16.0$ from a control of $84.6 \pm 23.9$) was not significantly different from that observed in response to the parasympathetic blockade ($-82.9 \pm 29.0$ from a control of $94.1 \pm 32.4$, $P=0.5$).

SAD did not significantly change both the number and the mean slope of the nonbaroreflex sequences (Table 2 and Figure 2). On the contrary, the number of lag 0 sequences was significantly decreased by SAD (Figure 3). As expected, the baroreflex sequences showed a significant and substantial decrease (Figure 4).

Controlled respiration did not significantly affect the number of the nonbaroreflex sequences (Figure 5). In this experimental set, we did not focus on baroreflex sequences because it has previously been shown that controlled respiration does not affect either their number or the mean slope.3

### Table 1. Cardiovascular and Respiratory Responses to Sympathetic Blockade, Atropine, Complete Autonomic Blockade, and SAD

<table>
<thead>
<tr>
<th></th>
<th>SAP, mm Hg</th>
<th>DAP, mm Hg</th>
<th>HR, bpm</th>
<th>V̇E, mL/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>132.2±4.3</td>
<td>81.3±2.0</td>
<td>319.7±8.9</td>
<td>874±28.4</td>
</tr>
<tr>
<td>Sympathetic blockade (n=15)</td>
<td>78.9±3.8‡</td>
<td>47.4±1.8‡</td>
<td>233.4±4.5‡</td>
<td>1205±85.4‡</td>
</tr>
<tr>
<td>Control</td>
<td>140.6±3.5</td>
<td>79.2±1.8</td>
<td>278.3±9.1</td>
<td>837±66.9</td>
</tr>
<tr>
<td>Atropine (n=16)</td>
<td>129.8±3.0*</td>
<td>73.9±2.3*</td>
<td>296.7±9.3*</td>
<td>1081±119.4*</td>
</tr>
<tr>
<td>Control</td>
<td>136.7±5.3</td>
<td>78.3±2.4</td>
<td>281.7±10.7</td>
<td>820±69.4</td>
</tr>
<tr>
<td>Complete autonomic blockade (n=13)</td>
<td>74.9±5.2‡</td>
<td>46.1±4.0‡</td>
<td>206.8±8.1‡</td>
<td>1359±148.0‡</td>
</tr>
<tr>
<td>Control</td>
<td>133.4±4.7</td>
<td>76.5±2.4</td>
<td>282.6±11.6</td>
<td>1023±123.0</td>
</tr>
<tr>
<td>SAD (n=10)</td>
<td>128.2±6.2</td>
<td>81.4±4.5</td>
<td>303.2±10.4*</td>
<td>1127±112.0</td>
</tr>
</tbody>
</table>

DAP indicates diastolic arterial pressure. Values are mean±SEM.

*P<0.05; †P<0.01; ‡P<0.001 vs control.

### Table 2. Changes in the Number of Sequences in Response to Sympathetic Blockade, Atropine, Complete Autonomic Blockade, and SAD

<table>
<thead>
<tr>
<th></th>
<th>$-\Pi/1 + SAP$, n</th>
<th>$+\Pi/2 - SAP$, n</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>32.4±11.9</td>
<td>52.2±14.2</td>
</tr>
<tr>
<td>Sympathetic blockade (n=15)</td>
<td>14.3±6.1*</td>
<td>11.0±6.1‡</td>
</tr>
<tr>
<td>Control</td>
<td>48.5±18.8</td>
<td>45.6±14.5</td>
</tr>
<tr>
<td>Atropine (n=16)</td>
<td>8.1±5.1*</td>
<td>3.1±2.1†</td>
</tr>
<tr>
<td>Control</td>
<td>42.0±19.0</td>
<td>46.7±17.9</td>
</tr>
<tr>
<td>Complete autonomic blockade (n=13)</td>
<td>1.2±0.8*</td>
<td>17.9±11.7*</td>
</tr>
<tr>
<td>Control</td>
<td>41.3±20.4</td>
<td>18.1±5.3</td>
</tr>
<tr>
<td>SAD (n=10)</td>
<td>32.5±15.7</td>
<td>27.8±9.4</td>
</tr>
</tbody>
</table>

Values are mean±SEM.

*P<0.05; †P<0.01; ‡P<0.001 vs control.
Discussion
The main goal of the present investigation was to test the hypothesis that nonbaroreflex sequences of spontaneously occurring consecutive heartbeats characterized by consensual, linearly related increases in SAP and HR (decrease in PI) could reflect a cardiovascular regulatory mechanism modulated by the autonomic nervous system operating with feed-forward (or positive feedback) characteristics.

Previous studies dealing with the possible role played by feed-forward mechanisms in the regulation of the cardiovascular system have addressed mainly the circulatory mechanisms modulating blood pressure and HR dynamics through the "runoff phenomenon" or the mechanical coupling between HR and arterial pressure. These mechanisms could have been involved in determining the occurrence of the nonbaroreflex sequences. We tried to disentangle the contribution of mechanical from putative neural feed-forward mechanisms by inducing a complete autonomic blockade at the target function level. Complete autonomic blockade drastically and significantly reduced the number of the hypertensive/tachycardic and hypotensive/bradycardic sequences, thus indicating that in anesthetized rabbits the occurrence of spontaneous nonbaroreflex sequences is mediated primarily by the autonomic nervous system.

We also tried to obtain a more detailed picture of the neural pathways involved in the occurrence of the nonbaroreflex sequences by inducing selective blockades of the efferent sympathetic and parasympathetic activities. Both sympathetic and parasympathetic blockades significantly decreased the number of "nonbaroreflex sequences, thus suggesting that both branches of the autonomic nervous system take part in this positive feedback mechanism of short-term cardiovascular neural regulation.

Our finding of a sympathetic contribution to the occurrence of nonbaroreflex sequences is consistent with that of Pagani et al of a positive feedback sympathetic reflex elicitable in the dog by mechanical stretching of the thoracic aorta. In that study, the consensual increases in SAP and HR induced by...
aortic distension (within ranges mimicking the effects of physiological increases in AP on aortic wall) were in fact reduced or abolished after pharmacological sympathetic blockade.

In our study, sympathetic blockade not only produced a decrease in the occurrence of nonbaroreflex sequences but also resulted in a significant increase in the gain of the spontaneous baroreflex sequences. An increased baroreflex sensitivity after sympathetic blockade has consistently been reported in humans,\(^9\,13\) but the mechanisms responsible for this enhancement have not been clearly established.\(^11\,14\) One possible explanation, ensuing from previous studies\(^6\,13\) and supported by the present one, is that the increased spontaneous baroreflex sensitivity observed after sympathetic blockade might be caused in part by the decreased influence of feed-forward neural mechanisms normally opposing the negative feedback mechanisms of baroreflex origin. Pagani et al\(^6\) reported an attenuation of the bradycardic response to phenylephrine during the hypertensive/tachycardic reflex elicited in the conscious dog by stretching of the thoracic aorta, an observation that is reciprocal of our result of an increased gain in the integrated spontaneous baroreceptor-cardiac reflex after sympathetic blockade. On the other hand, the interruption of the sympathomediated excitatory influences by activation of cardiovascular spinal afferents has been reported to increase the reflex bradycardic response to acute arterial pressure rises.\(^15\)

Takken together, the results of previous\(^6\,15\) and present investigations support the concept that neural regulatory mechanisms with positive feedback characteristics play a role in the physiological control of circulation interacting dynamically with the negative feedback mechanisms.\(^7\)

It is noteworthy that both the hypertensive/tachycardic and hypotensive/bradycardic sequences appear to be modulated by the sympathetic nervous system inasmuch as both decreased significantly after sympathetic blockade. The complex nature of neural cardiovascular regulation is further stressed by the observation that the occurrence of the nonbaroreflex sequences appears to be also under the control of the parasympathetic branch of the autonomic nervous system, as indicated by their significant decrease after atropine alone. Overall, the above results might be interpreted as indicating that the occurrence of nonbaroreflex sequences is due to a synergistic contribution of the sympathetic and parasympathetic activity, whose relative modalities are difficult to ascertain. Growing evidence indicates that the sympathetic and parasympathetic nervous systems may regulate the cardiovascular system in a parallel instead of a reciprocal fashion,\(^16\,19\) interacting at different levels.\(^20\,21\) On the other hand, physiological variations in autonomic outflows have been shown to be capable of modulating the expression of feed-forward regulatory mechanisms in humans.\(^9\,22\)

The mean slope of the nonbaroreflex sequences that within the framework of this study would represent the approximate gain in the transfer function of the positive feedback control mechanisms was not significantly changed by pharmacological manipulations of the autonomic nervous system. At variance with the baroreflex sequences that are mainly vagally mediated,\(^2\,23\) the nonbaroreflex sequences appear to be modulated by both the parasympathetic and sympathetic nervous systems. How the nonlinear frequency properties of these components and their dynamic interaction\(^17\) may have affected the gain of the nonbaroreflex sequences cannot be elucidated with the sequences technique. Further studies incorporating more complex methodologies are needed to clarify this point.

Our data indicate that the lag used to pair SAP and PI values significantly affects the physiological meaning of sequences of spontaneously occurring consecutive heartbeats. Like the nonbaroreflex (ie, lag 0) sequences, lag 0 sequences would be neurally mediated because of their decrease after complete autonomic blockade. However, at variance with the nonbaroreflex, lag 0 sequences appear to be modulated by the arterial baroreflexes because they were significantly reduced by SAD, which did not affect lag 1 sequences. These results would confirm those of Blaber et al\(^22\) suggesting that consecutive heartbeat sequences in which the SAP and RR interval of the same beat change in the opposite direction could represent artifacts created by baroreflex activity related to a phase relationship between systolic blood pressure and RR interval.\(^24\)

We used controlled breathing to determine whether respiration, with its neural and nonneural modulating effects on arterial pressure and HR,\(^25\,26\) was linked to the observation of nonbaroreflex sequences. However, the more regular breathing occurring during artificially induced ventilation did not significantly affect the occurrence of nonbaroreflex sequences (Figure 5).

**Study Limitations**

The main limitation is that only a small number of beats (≈5%) was organized in sequences characterized by a nonbaroreflex pattern. This finding should not be interpreted as indicating that feed-forward mechanisms are not engaged for a prominent fraction of time or that they modulate the sinus node only in a sequencelike fashion. It may only reflect an intrinsic limitation of the sequences method. This observation could be also explained, at least in part, by the use of anesthetized animals because acute anesthesia negatively affects the cardiovascular neural modulation. The small value of the slopes observed in the present study also might have been caused in part by the anesthesia and the elevated level of HR.\(^23\,27\) However, these factors did not prevent us from observing significant effects on spontaneous baroreflex slope when they occurred, such as after sympathetic blockade. We also considered the possibility that the anesthetic agents could have abolished some neural traffic, so that by the time the pharmacological blockades of the autonomic nervous system were done, there was not much efferent sympathetic and/or parasympathetic activity to oppose. However, the significant cardiovascular changes in response to the different pharmacological blockades (Table 1) make this possibility unlikely, suggesting that despite the anesthesia, there was enough sympathetic and/or parasympathetic activity to oppose in the experimental conditions of the present investigation.

In conclusion, the results of this study suggest that the continuous relationship between spontaneous fluctuations in arterial blood pressure and heart period is neurally modulated.
not only through negative feedback coupling mechanisms, as reflected by the baroreflex sequences, but also through positive feedback mechanisms, as would be reflected by the nonbaroreflex sequences characterized by consensual, linearly related changes in SAP and HR. Finally, this neurally mediated, feed-forward type of short-term cardiovascular regulation seems able to interact dynamically with the negative feedback mechanisms in the control of heart period.

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