Baroreceptor mechanisms controlling sympathetic nervous rhythms of central origin

DAVID G. TAYLOR AND GERARD L. GEBBER
Department of Pharmacology, Michigan State University, East Lansing, Michigan 48824

TAYLOR, DAVID G., AND GERARD L. GEBBER. Baroreceptor mechanisms controlling sympathetic nervous rhythms of central origin. Am. J. Physiol. 228(4): 1002-1013. 1975.—A study was made in the anesthetized cat of the slow wave of sympathetic nervous discharge (SND) locked in a 1:1 relation to the cardiac cycle (3 cycle/s periodicity). SND was recorded from the preganglionic splanchnic and postganglionic renal nerves. The data contradict the generally accepted view that the slow wave occurs as the direct result of a waxing and waning of baroreceptor nervous discharge. Although baroreceptor denervation unlocked the phase relations between SND and the cardiac cycle, the slow wave persisted and its duration was not changed. Furthermore, the slow wave was aborted by stimuli delivered to the baroreceptor nerves or paramedian reticular nucleus during a time span which accounted for less than 1% of the cardiac cycle. It is concluded that the 3 cycle/s periodicity of SND is representative of a vasomotor rhythm of central origin which is entrained to the cardiac cycle by the baroreceptor reflexes. The sympathetic inhibitory effect leading to the entrainment of the slow wave is mediated in the brainstem. A spinal component of baroreceptor-induced sympathoinhibition also was demonstrated.

SYMPATHETIC NERVOUS RHYTHMS OF CENTRAL ORIGIN

Oscillations of sympathetic nervous discharge (SND) locked in a 1:1 relation to the cardiac cycle (~3 cycle/s periodicity) are generally considered to result from a waxing and waning of baroreceptor nervous discharge associated respectively with the systolic and diastolic phases of the arterial pulse (1, 8, 20–21). This is supported by the observation that the phase relations between SND and the cardiac cycle disappear after interruption of baroreceptor nervous discharge (1, 3, 6, 12, 13, 19, 20, 30, 35, 37). However, while some investigators (3, 6, 37) reported that SND appeared essentially continuous or random in character following removal of baroreceptor nervous discharge, others (2, 13, 30) observed irregularly occurring oscillations of SND indicative of synchronized activity of individual fibers contained within preganglionic nerve bundles. The records presented by Downing and Siegel (13) and Koizumi et al. (30) are particularly intriguing since the duration of the synchronous bursts of SND appears to approach that of those which were locked to the cardiac cycle before section of the baroreceptor nerves. The work of Kezdi and Geller (29) also is pertinent. They observed the effects produced on renal SND by nonpulsatile and pulsatile pressure applied to the isolated catod sinus of the dog. The aortic depressor nerves were cut in these experiments. Figure 1 of their study shows synchronous bursts of SND when nonpulsatile pressure was applied to the carotid sinus. This is striking since the discharge of individual baroreceptor fibers is uniform rather than rhythmic under these conditions (14, 20). These observations raise the possibility that oscillations of SND which appear locked in a 1:1 relation to the cardiac cycle are not generated by the baroreceptor reflexes. Rather, the 3 cycle/s periodicity of SND may be representative of a vasomotor rhythm of central origin which is entrained by the baroreceptor reflexes. The present investigation was designed to test this hypothesis.

METHODS

Sixty-seven cats weighing between 2.0 and 3.5 kg were anesthetized by the intraperitoneal injection of a mixture of sodium diallylbarbiturate (70 mg/kg), urethan (280 mg/kg), and monoethylnurea (280 mg/kg). Rectal temperature was maintained between 36 and 38°C with a heating pad and lamp. The animals were immobilized with gallamine triethiodide (4 mg/kg, iv) and artificially respired. Supplemental doses (2 mg/kg, iv) of gallamine were administered as required during the course of the experiment to prevent somatomotor responses to stimulation of the neuraxis. Pneumothoracotomy was performed to minimize movements associated with artificial respiration. Blood pressure was monitored from the tip of a catheter inserted into the lumbar aorta via the femoral artery and displayed on a Grass polygraph (model 7B).

Baroreceptor nerve section and stimulation. The carotid sinus, aortic depressor and vagus nerves were exposed from a ventral aspect after reflection of a portion of the trachea and esophagus into the mouth. The carotid sinus nerve was isolated at its junction with the glossopharyngeal nerve, while the aortic depressor nerve was located at its junction with the superior laryngeal nerve. The vagus was isolated in the midecervical region. Baroreceptor denervation was accomplished by bilateral section of these nerves. The central ends of the baroreceptor nerves were placed on bipolar platinum electrodes and stimulated with square-wave pulses.

Central stimulation and histology. The cat was placed in a David Kopf stereotaxic apparatus and a portion of the occipital bone overlying the caudal medulla was removed. The cerebellum was removed by gentle suction in some experiments. The dorsal portions of the first, second and fourth cervical vertebrae were also removed. Stimuli were
BARORECEPTORS AND SYMPATHETIC RHYTHMS

passed from a Grass S88 stimulator through a stimulus-isolation unit to bipolar concentric stainless steel electrodes placed at selected sites in the medulla and spinal cord. Square-wave pulses were of supramaximal intensity (10 V) with durations of 0.5 ms. Stimulation was performed on the side ipsilateral to the recording electrodes on the splanchnic and renal sympathetic nerves. The center lead of the stimulating electrode and the shaft (outer contact) were exposed for 0.25 mm and separated by 0.5 mm. The electrodes were stereotaxically positioned in the medulla according to the coordinates of Berman (4).

The medulla and spinal cord were removed and fixed in Formalin at the end of each experiment. Sections of 30 μm thickness were cut with a cryostat-microtome and stained with cresyl violet. Sections were cut in the frontal plane for the identification of electrode tracks.

Sympathetic nerve recording and data analysis. The left pre-ganglionic greater splanchnic nerve and postganglionic renal nerves were exposed via a retroperitoneal approach. The splanchnic nerve was identified in the area of the costovertebral triangle and cut at its entrance into the celiac ganglion. One of the renal nerves was traced to and sectioned near its entrance into the kidney. Nerve potentials were recorded monophasically under oil with bipolar platinum electrodes after capacity-coupled preamplification (low and high half-amplitude responses at 1 and 1,000 Hz). Nerve activity was stored on magnetic tape and simultaneously displayed on a polygraph. The time relations between SND and the R wave of the ECG, or electrical stimuli applied to the neuraxis and baroreceptor nerves, were analyzed by computer summation (Nicolet model 1070).

The memory content of the computer was displayed in analog form on an oscilloscope (Tektronix model 502), or X-Y recorder. The sweep of the computer was initiated by a SYNC pulse from the stimulator. The SYNC pulse was generated in conjunction with square-wave pulses applied to the neuraxis or baroreceptor nerves, or by a timing pulse from a logic-divider trigger circuit which was derived from the R wave of the ECG. It was possible to apply stimuli to the neuraxis at any desired point in the cardiac cycle by interfacing the logic divider trigger circuit with the delay circuit of the stimulator. The autocorrelation function of splanchnic and renal nerve activity was also analyzed in some experiments with the Nicolet computer.

Statistical analysis. Statistical analysis was performed with the Student t test for paired and unpaired data. P values of < 0.05 were considered to indicate statistical significance. Values are expressed as means ± SE.

RESULTS

Figure 1A shows the characteristics of a spontaneous burst of renal nerve activity recorded simultaneously with bandpass settings of 1–1,000 and 30–1,000 Hz. With the high-pass filter set at 1 Hz, SND was viewed primarily as a slow wave, the characteristics of which will be the subject of this paper. The high-frequency components which formed the slow wave are shown in the lower record of Fig. 1A (high-pass filter set at 30 Hz). Note that the duration of the slow wave accurately depicts the period of high-frequency spiking. These records support the contention of Cohen and Gootman (8) that slow waves of SND result from synchronized activity (spikes) of individual fibers contained within the nerve bundle under study. That is, the slow wave represents an envelope of nerve spikes. Figure 1, B–C, shows that the rise in systemic blood pressure produced by the intravenous injection of 1 μg/kg of norepinephrine (baroreceptor reflex activation (16)) was associated with a disappearance of the slow waves. This observation further attests to the neural origin of the slow wave.

Is the 3 cycle/s periodicity of SND generated or entrained by baroreceptor reflexes? Computer summation of 64 sweeps triggered by a timing pulse derived from the R wave of every fourth ECG complex enabled us to analyze the phase rela-
Figure 2. Relationship between cardiac cycle and SND in 2 cats with intact baroreceptor reflexes. A: computer-summed traces (64 sweeps) of arterial pulse (top) and splanchnic or renal SND (bottom). Computer sweep was triggered by R wave of every 4th ECG complex. Mean blood pressure was 110 mmHg (splanchnic) and 107 mmHg (renal). B: autocorrelation functions of splanchnic and renal SND. Address bin was 4 ms. Sweep duration was 1 s. Sample run was 4 min. Horizontal calibration is 500 ms. Vertical calibration is 133 µV and refers to records in A.

Figure 3 demonstrates that the slow wave of SND persisted after section of the baroreceptor and vagus nerves (four experiments) or hemorrhage (six experiments) to a mean arterial pressure between 70 and 80 mmHg (HEMOR I). However, the phase relations between SND and the cardiac cycle were unlocked (Fig. 4). This was shown when the oscillographic records from each experiment were subjected to computer summation. The records in Fig. 4 were derived, in part, from the oscillographic tracings illustrated in Fig. 3, A and B (HEMOR I). Computer-summed records of SND triggered by the R wave after baroreceptor denervation or hemorrhagic approached a straight line since signal averaging reduces those components of the electrical recording which are not time-locked to the cardiac cycle in proportion to the square root of the number of trials summed. These records were indistinguishable from those of summed traces triggered by dummy pulses applied randomly with respect to the cardiac cycle. This observation attests to the complete removal of baroreceptor nervous input. Bilateral section of the carotid sinus, aortic depressor and vagus nerves also eliminated inhibition of SND associated with the pressor action of intravenous norepinephrine. The autocorrelation

SND depicted as slow waves on oscillographic records when the high-pass filter was set at 1 Hz.

Others (13, 30) have shown that SND is not strictly uniform or continuous in character following removal of baroreceptor reflex influences. However, the high-pass filter settings (30–80 Hz) used in these studies would have made it difficult to measure and compare sympathetic nervous wave forms before and after baroreceptor denervation (see lower trace of Fig. 1A). In contrast, we were able to measure accurately the characteristics of synchronized bursts of SND described as slow waves on oscillographic records when the high-pass filter was set at 1 Hz.
the slow wave often was more prominent after removal of dure. However, high-frequency spiking superimposed on denervation or when bleeding was carried to the point of occurrence of the slow wave was significantly increased by baroreceptor nervous input (Fig. 8C).

Table I further supports this contention. The negative phase of the slow wave of renal nerve activity was not significantly changed by baroreceptor denervation or hemorrhage. The amplitude of the slow wave was not significantly affected by either procedure. Most importantly, the duration (~200 ms) of the negative phase of the slow wave of renal nerve activity was not significantly changed by baroreceptor denervation or hemorrhage. In addition, Table I shows that the frequency of occurrence of the ~200-ms slow wave of SND. This is shown in the lower traces of Fig. 3B (HEMOR II). The slow wave was replaced in large part by a wave form with a duration of approximately 100 ms. Thus, severe hemorrhage but not baroreceptor denervation led to partial "desynchronization" of SND. That is, large-amplitude slow waves were replaced by higher frequency activity.

The results presented in Figs. 3 and 4 suggest that the slow wave was generated by central vasomotor elements, rather than by the baroreceptor reflexes as has been generally assumed (1, 8, 21). Table I further supports this contention. Most importantly, the duration (~200 ms) of the negative phase of the slow wave of renal nerve activity was not significantly changed by baroreceptor denervation or hemorrhage. In addition, Table I shows that the frequency of occurrence of the slow wave was significantly increased by baroreceptor denervation or hemorrhage. The amplitude of the slow wave was not significantly affected by either procedure. However, high-frequency spiking superimposed on the slow wave often was more prominent after removal of baroreceptor nervous input (Fig. 8C).

In contrast to the results observed following baroreceptor denervation or when bleeding was carried to the point of functions of SND in Fig. 4 show a reduced degree of periodicity after section of the baroreceptor nerves or hemorrhage. This observation indicates that the slow waves of SND were less evenly spaced following the removal of baroreceptor nervous input.

The results presented in Figs. 3 and 4 suggest that the slow wave was generated by central vasomotor elements, rather than by the baroreceptor reflexes as has been generally assumed (1, 8, 21). Table I further supports this contention. Most importantly, the duration (~200 ms) of the negative phase of the slow wave of renal nerve activity was not significantly changed by baroreceptor denervation or hemorrhage. In addition, Table I shows that the frequency of occurrence of the slow wave was significantly increased by baroreceptor denervation or hemorrhage. The amplitude of the slow wave was not significantly affected by either procedure. However, high-frequency spiking superimposed on the slow wave often was more prominent after removal of baroreceptor nervous input (Fig. 8C).

In contrast to the results observed following baroreceptor denervation or when bleeding was carried to the point of

![B] CONTROL [DENERVATED]

![A] CONTROL [HEMORRHAGE]

![C] CONTROL [HEMORRHAGE]

![D] CONTROL [HEMORRHAGE]

![E] CONTROL [HEMORRHAGE]

![F] CONTROL [HEMORRHAGE]

![G] CONTROL [HEMORRHAGE]

![FIG. 4. Unlocking of phase relations between SND and cardiac cycle. Records are computer readouts of oscillographic tracings shown in Fig. 3. A: baroreceptor denervation (bilateral section of carotid sinus, aortic depressor, and vagus nerves). A, 1 and 2: computer-summed traces (64 sweeps) of arterial pulse (1) and renal SND (2) before and after baroreceptor denervation. Computer sweep was triggered by R wave of every 4th ECG complex. Changes in mean blood pressure are not depicted in computer readouts of the arterial pulse. A3: sum of 64 computer sweeps triggered by dummy pulses applied randomly with respect to cardiac cycle. A4: autocorrelation functions of SND before and after baroreceptor denervation. Address win was 10 ms. Sweep duration was 2.5 s. Sample run was 5 min. B: hemorrhage to lower mean blood pressure from 112 to 72 mmHg. B1: computer-summed slow wave. When the single shock was applied close to the beginning of or near peak systole, the first slow wave of the triplet was extinguished or prematurely terminated the slow wave of SND. Figure 5 is typical of the nine experiments performed. The computer-summed records in Fig. 5A show locking of the slow wave of renal nerve activity to the arterial pulse. Panels B-G compare the 3 cycle/s oscillation of SND shown in panel A with the computer-summed wave forms following the application of a single shock to the paramedian nucleus at selected points in the cardiac cycle. Peak amplitude of the first slow wave in the triplet was reduced and delayed in time when the stimulus was applied simultaneously with the timing pulse (derived from R wave of ECG) which triggered the sweep of the computer (panel B). The original oscillographic records revealed that the first slow wave was extinguished approximately 50% of the time by the stimulus applied to the paramedian nucleus. This most likely accounted for the reduction in amplitude of the computer-summed slow wave. When the single shock was applied close to the beginning of or near peak systole, the first slow wave of the triplet was extinguished or prematurely terminated (panels C and D), respectively. Application of the stimulus in early, mid, or late diastole failed to affect the slow wave (panels E-G). The record in panel E is particularly interesting since it shows that the stimulus was ineffective when placed approximately 60 ms after the start of the slow wave. Neither a facilitatory nor an occlusive interaction

TABLE 1. Effect of baroreceptor denervation and hemorrhage on characteristics of slow wave of renal SND (3 cycle/s periodicity) measured directly from oscillographic tracings

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Control</th>
<th>Postdenervation</th>
<th>%Δ*</th>
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<tbody>
<tr>
<td><strong>A) Baroreceptor denervation, n = 4</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Duration, ms†</td>
<td>211 ± 7</td>
<td>203 ± 6, -5 ± 7</td>
<td></td>
</tr>
<tr>
<td>Frequency of occurrence, cycles/min</td>
<td>178 ± 13</td>
<td>197 ± 13, +11 ± 18</td>
<td></td>
</tr>
<tr>
<td>Amplitude, μV</td>
<td>35 ± 10</td>
<td>36 ± 12, -1 ± 9</td>
<td></td>
</tr>
<tr>
<td><strong>B) Hemorrhage, ‡ n = 6</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Duration, ms†</td>
<td>191 ± 7</td>
<td>191 ± 6, +1 ± 5</td>
<td></td>
</tr>
<tr>
<td>Frequency of occurrence, cycles/min</td>
<td>186 ± 7</td>
<td>202 ± 7, +12 ± 48</td>
<td></td>
</tr>
<tr>
<td>Amplitude, μV</td>
<td>38 ± 7</td>
<td>32 ± 14, +37 ± 29</td>
<td></td>
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</tbody>
</table>

Values are means ± SE calculated from 10-60 consecutively occurring slow waves. *Mean ± SE calculated from percent change measured in each experiment. † Duration of negative phase of slow wave. ‡ Hemorrhage to a mean arterial pressure (70-80 mmHg) at which the phase relations between SND and the cardiac cycle were unlocked. § Statistical significance at the P < 0.05 level (paired comparison).
FIG. 5. Effect of single shocks (10 V, 0.5 ms) applied to paramedian reticular nucleus on slow wave of renal SND. A: computer-summed traces (64 sweeps) of arterial pulse (top) and SND (bottom). Computer sweep was triggered by R wave of every 4th ECG complex. Mean blood pressure was 135 mmHg. B-G: comparison of computer summed trace of SND shown in A (dotted line) with those (solid line) summed following paramedian stimulation at selected points in cardiac cycle. Single shocks applied to the paramedian nucleus were delayed as follows with respect to the R wave. B: 0 ms; C: 50 ms; D: 120 ms; E: 150 ms; F: 200 ms; G: 250 ms. Arrows show point of application of single shock. Horizontal calibration is 500 ms. Vertical calibration is 267 μV.

FIG. 6. Computer-summed traces (64 sweeps) of splanchnic and renal nerve positive potentials evoked by stimulation of paramedian reticular nucleus. A single shock or a 5-ms train of three pulses (10 V) applied once every 4 s to depressor sites in the paramedian nucleus. The stimuli were applied randomly with respect to the cardiac cycle. Inhibition of spontaneously occurring SND was displayed as a positive wave by the computer. Positive potentials result from intervals of decreased SND time-locked to the stimulus which average as periods of lesser negativity than are occurring spontaneously (7, 18, 42). Although single shocks or trains of pulses applied to the paramedian nucleus failed to change blood pressure, stimulation at 50 Hz for 15 s decreased mean arterial pressure by 30–80 mmHg.

Figure 6 illustrates the positive potentials evoked in the splanchnic or renal sympathetic nerves of four different cats. Stimulation of the paramedian nucleus with a single shock evoked a wave of positivity which appeared similar in form to the spontaneously occurring slow negative wave of SND. In addition, an earlier positive potential was elicited by trains of pulses applied to the paramedian nucleus. The early positivity was considerably shorter in duration than the late positive potential. The time course of the early and late positive potentials often overlapped (Fig. 6, B, C, and D). The duration of the late positive potential was essentially between naturally occurring baroreceptor nervous and electrically evoked activity was important in determining the effectiveness of paramedian stimulation. This is shown by the traces in panels C and D. In panel C, the single shock was applied at a time when naturally occurring baroreceptor nervous activity would have been at a minimum (pre-systole). In panel D, the stimulus was applied near peak systole, i.e., during a time when naturally occurring baroreceptor nervous activity would have been maximal. Importantly, the stimulus was effective in terminating the slow wave in both cases.

The data in Fig. 5 also make it difficult to envision how the slow wave could result from a waxing and waning of baroreceptor nervous discharge occurring during each cardiac cycle. It is reasonable to assume that the effects of paramedian stimulation monitored activation of intramedullary components of the baroreceptor reflex arc since the carotid sinus nerve makes primary and secondary connections with this medial medullary nucleus (11, 22, 23, 32, 33). In addition, stimulation of the paramedian nucleus mimics the effects of baroreceptor reflex activation on pre- and postganglionic SND (40, 41). This assumption will be substantiated in a subsequent section of the RESULTS.

Temporal characteristics of sympathoinhibition evoked by stimuli applied to paramedian nucleus. The results presented in Fig. 5 led to the prediction that the time course of computer-summed inhibition of SND induced by paramedian stimulation would be similar to that of the spontaneously occurring slow negative wave. This was tested by summing 64 splanchnic or renal nerve responses produced by single shocks or 5-ms trains of three pulses (10 V) applied once every 4 s to depressor sites in the paramedian nucleus. The stimuli were applied randomly with respect to the cardiac cycle. Inhibition of spontaneously occurring SND was displayed as a positive wave by the computer. Positive potentials result from intervals of decreased SND time-locked to the stimulus which average as periods of lesser negativity than are occurring spontaneously (7, 18, 42). Although single shocks or trains of pulses applied to the paramedian nucleus failed to change blood pressure stimulation at 50 Hz for 15 s decreased mean arterial pressure by 30–80 mmHg.
the same, independent of whether it was elicited by a single shock or a train of pulses. The temporal characteristics of the early and late positive potentials are summarized in Table 2.

Figure 7 shows that the late positive potential evoked by

**Figure 7.** Comparison of time course of late positive potential (renal nerve) evoked by stimulation of paramedian reticular nucleus with that of slow wave of SND derived in same cat by computer summation and autocorrelation analysis. A: superimposition of traces shown in B-D; positivity (dotted line) is inverted; computer-summed slow wave (solid line); one cycle of autocorrelation function (dot-dash line). B: computer-summed positivity (64 sweeps) evoked by single shock (10 V, 0.5 ms) applied to paramedian nucleus randomly with respect to cardiac cycle. C: computer-summed traces (64 sweeps) of arterial pulse (top) and SND (bottom). Computer sweep was triggered by R wave of every fourth ECG complex. D: autocorrelation function of SND. Address bin was 4 ms. Sweep duration was 1 s. Sample run was 4 min. Horizontal calibration is 133 μV for B and C.

**TABLE 2.** Temporal characteristics of early and late positive potentials evoked by 3-ms trains of three pulses applied to paramedian nucleus

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Splanchnic Nerve</th>
<th>Renal Nerve</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Early +</td>
<td>Late +</td>
</tr>
<tr>
<td>Onset latency, ms</td>
<td>28 ± 3 (22)</td>
<td>99 ± 8 (22)</td>
</tr>
<tr>
<td>Duration, ms</td>
<td>72 ± 11 (12)*</td>
<td>294 ± 22 (22)</td>
</tr>
</tbody>
</table>

Values are means ± SE with (n). * Measurements from experiments in which early and late positive potentials did not overlap.

**TABLE 3.** Comparison of temporal characteristics of computer-summed late positivity and spontaneously occurring slow wave of SND recorded from renal nerve in 12 cats

<table>
<thead>
<tr>
<th></th>
<th>Time-to-Peak Amplitude, ms</th>
<th>Total Duration, ms</th>
</tr>
</thead>
<tbody>
<tr>
<td>Late positivity</td>
<td>85 ± 6</td>
<td>209 ± 9</td>
</tr>
<tr>
<td>Slow wave of SND</td>
<td>94 ± 6</td>
<td>211 ± 11</td>
</tr>
</tbody>
</table>

Values are means ± SE.
FIG. 8. Relationships between character of spontaneously occurring SND and positive potentials evoked by stimulation of paramedian reticular nucleus. Records in A, B, and C are from 3 different cats. A: effect of severe hemorrhage to a mean arterial pressure of 55 mmHg on splanchnic nerve activity; 1, oscillographic tracings of SND before and after hemorrhage; 2, computer-summed traces (32 sweeps) of positivity evoked by a 5-ms train of 3 pulses applied to paramedian nucleus randomly with respect to cardiac cycle before and after hemorrhage. B: effect of asphyxia (15-47 s after artificial respirator was turned off) on renal nerve activity; 1 and 2, as described for A. C: effect of baroreceptor denervation (bilateral section of carotid sinus, aortic depressor, and vagus nerves) on renal nerve activity; 1 and 2, as described for A. Horizontal calibrations are 500 ms. Vertical calibrations are 20 PV for oscillographic tracings and 267 PV for records of positivity.

Response patterns are depicted in Fig. 9. Each response pattern was observed at least 2 times for each of the two baroreceptor nerves stimulated (17 experiments). Most importantly, both early and late positive potentials could be evoked independently of preceding negativity. The temporal characteristics of the positive potentials were similar to those reported in Table 2 for positivity produced by stimulation of the paramedian nucleus. Thus two distinct phases of sympathoinhibition were associated with baroreceptor nerve stimulation.

Brainstem and spinal components of baroreceptor-induced sympathoinhibition. The question arose concerning the level(s) of the neuraxis at which the early and late phases of baroreceptor-induced sympathoinhibition were mediated. The data illustrated in Fig. 10 are representative of those obtained in four cats in which the effect of midcollicular transection was tested on the positive potentials elicited in the splanchnic nerve by stimulation of the aortic depressor nerve (panel A) and the paramedian nucleus (panel B). Decerebration performed according to the method of Gebber et al. (16) had little effect on the early positive potential, but usually prolonged the duration and sometimes enhanced.
the amplitude of the late positive potential. The onset latency of the late positive potential was not significantly changed. Although not shown in Fig. 10 the slow wave of SND persisted after decerebration. These experiments suggest that sympathoinhibition evoked by stimulation of the aortic depressor nerve and paramedian nucleus occurred below the midscollicular level and did not require forebrain loops for its initiation.

The effects of baroreceptor reflex activation and paramedian stimulation were tested on potentials evoked in the splanchnic nerve by 10-ms trains of three pulses (10 V) applied once every 2 s to dorsolateral medullary pressor sites, and to descending pressor tracts in the dorsolateral white column of the fourth cervical spinal segment (24, 25, 27, 28). The medullary and spinal pressor regions explored have been delineated in previous reports from this laboratory (16, 40, 41). Sixteen splanchnic nerve potentials were summed before and during inhibition of spontaneously occurring SND associated with 1) the pressor action produced by the intravenous injection of 1 μg/kg of norepinephrine (i.e., baroreceptor reflex activation (16)), and 2) the depressor effect produced by paramedian stimulation (10 V, 0.5 ms, 50 Hz). The summed splanchnic nerve potentials evoked from medullary pressor sites were inhibited to a significantly greater degree than those evoked from spinal pressor sites (Fig. 11 and Table 4). This observation supports the contention of Gebber et al. (16) that baroreceptor-induced sympathoinhibition occurs both at spinal and brainstem levels. Two observations indicated that the splanchnic nerve potentials evoked from the dorsolateral white columns of the midcervical spinal cord monitored the activation of descending pressor tracts. First, the onset latency of the splanchnic potential elicited from spinal pressor sites was shorter than for the potential evoked from the medulla (Fig. 11). Second, C1 transection failed to abolish the splanchnic nerve responses evoked from spinal pressor sites (Fig. 11 and Table 4). Baroreceptor reflex activation or paramedian stimulation reduced the splanchnic nerve responses evoked from spinal pressor sites to a significantly greater extent than did C1 transection (Table 4).

FIG. 11. Effect of baroreceptor reflex activation (norepinephrine, 1 μg/kg, i.v.) or paramedian stimulation (10 V, 0.5 ms, 50 Hz) on splanchnic nerve response evoked by stimulation of a medullary and a spinal pressor site in same cat. A1: computer-summed traces (16 sweeps) of SND evoked by a 10-ms train of 3 pulses (10 V, 0.5 ms) applied once every 2 s to a pressor site in dorsolateral medullary reticular formation. A2: same, but during peak pressor action of norepinephrine. A3: same, but during paramedian stimulation. B1: computer-summed traces (16 sweeps) of SND evoked by stimulation of a pressor site in dorsolateral white column of 4th cervical spinal segment. B2 and B3: as described for A2 and A3. Post-C1: response evoked from spinal pressor site 10 min after C1 transection. Horizontal calibration is 100 ms. Vertical calibration is 133 μV.

from spinal pressor sites to a significantly greater extent than did C1 transection (Table 4).

Figure 12 illustrates two experiments in which the time course of splanchnic nerve positivity was compared with that of spinal inhibition produced by stimulation of a paramedian depressor site with a 5-ms train of three pulses. The time course of spinal inhibition is depicted by the excitability-recovery curve of the splanchnic nerve discharge (sum of 16 trials) elicited by a 10-ms train of three pulses applied to a pressor site in the dorsolateral white column of the midcervical spinal cord. The onset latencies of the computer-summed splanchnic nerve discharges were 33 ms in Fig. 12A and 32 ms in Fig. 12B. This indicated that the potentials were evoked from descending spinal tracts. The time course of depression of the splanchnic nerve potential evoked from the spinal cord followed only that of the early phase of positivity in both experiments. This observation suggests that the early phase of baroreceptor-induced sympathoinhibition occurred in the spinal cord while the late phase was mediated at a supraspinal level.

**DISCUSSION**

The data presented in this study contradict the generally accepted view that the slow wave of SND (3 cycle/s periodicity) results directly from a waxing and waning of baroreceptor nervous discharge occurring during each cardiac cycle (1, 8, 20, 21). Although baroreceptor denervation or hemorrhage unlocked the phase relations between SND and the cardiac cycle, the slow wave persisted and its duration was not changed. This observation supports the contention that the 3 cycle/s periodicity of SND was generated by a central vasomotor oscillator rather than directly by the baroreceptor reflexes. This was further indicated by experiments in which the slow wave was aborted by stimuli delivered to the baroreceptor nerves or to the paramedian nucleus during a time span which accounted for less than 1 % of the cardiac cycle. First, a single shock applied to the paramedian nucleus at appropriate points in the cardiac cycle extinguished or prematurely terminated the slow wave. The paramedian nucleus of the medial medulla functions as a relay station in the baroreceptor reflex arc (11, 22, 29, 32, 33, 40, 41). Second, the late positive potential produced by

**TABLE 4. Effect of baroreceptor reflex activation (BRA) and stimulation of paramedian nucleus on computer summed splanchnic nerve discharges evoked by 10-ms trains of three pulses applied to medullary and midcervical spinal pressor sites**

<table>
<thead>
<tr>
<th>Source</th>
<th>%Δ in Responses Evoked From</th>
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<tbody>
<tr>
<td>Medulla (42-83)</td>
<td>Spinal cord (35-45)</td>
</tr>
<tr>
<td>BRA*</td>
<td>-97 ± 3 (12)</td>
</tr>
<tr>
<td>Paramedian stimulation</td>
<td>-93 ± 4 (13)</td>
</tr>
<tr>
<td>C1 transection</td>
<td>-40 ± 9 (10)</td>
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</tbody>
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Values are means ± SE with (n). * Induced by raising systemic blood pressure (intravenous injection of 1 μg/kg of norepinephrine). a,b,c,d Significantly different from effect produced on splanchnic nerve responses evoked from medulla and spinal cord, respectively, is given in parentheses. e Significantly different from effect produced either by BRA or paramedian stimulation.
importantly, it appears that the bursts of SND associated with lower frequencies of sinusoidal pressure wave application to the isolated carotid sinus of the dog produced a decrease in the duration of the burst of SND perhaps related to a change in the number of 100-ms packets accompanying each pressure cycle rather than to shortening of the 3 cycles/s slow wave which seemed to be absent in Fig. 6 of Kezdi and Geller’s study.

Cohen and Gootman (7, 8) and Gootman and Cohen (17, 18) noted a well defined 10 cycle/s periodicity of SND on the cat splanchnic nerve which often was locked in a 3:1 relation to the cardiac cycle. Assuming that oscillation of SND locked in a 1:1 relation to the cardiac cycle was of baroreceptor origin, Cohen and Gootman (8) concluded that the 10 cycle/s periodicity represented the fundamental rhythmicity of the baroreceptor nerves. This view is open to question since it is clear from the present study that SND can also be synchronized at a frequency approximating 3 cycles/s after baroreceptor denervation. Thus, it seems more likely that the central vasomotor oscillator can synchronize SND over a range of frequencies (3–10 cycles/s). The predominant frequency presumably is determined by the existing experimental conditions. In this study, the 3 cycle/s periodicity of SND was predominant except under the conditions of severe hemorrhage and asphyxia (Figs. 3 and 8).

It is well established that an inverse relationship exists between the degree of activity recorded from baroreceptor and sympathetic nerves (12, 20, 29, 34). The present study indicates that the baroreceptor reflexes also function to entrain the slow wave of SND to the cardiac cycle. This was made clear by the observation that baroreceptor denervation led to uncoupling of the phase relations between the slow wave and the cardiac cycle. The slow waves of SND were less evenly spaced in the absence of baroreceptor nervous discharge. This was indicated by the reduced degree of periodicity in the autocorrelograms of SND after baroreceptor nerve section or hemorrhage. Thus entrainment of the slow wave by the baroreceptor reflexes regularized the 3 cycle/s rhythm of central origin. The frequency of occurrence of the slow wave of SND was significantly increased by baroreceptor denervation. Thus entrainment of the slow wave to the cardiac cycle by the baroreceptor reflexes also is frequency-limiting in effect.

The effectiveness of paramedian stimulation in terminating the slow wave was dependent on the point at which the single shock was applied (Fig. 5). A single shock extinguished or prematurely terminated the slow wave when delivered just before or at its onset. However, a single shock applied just after the beginning of the slow wave had no effect. This could not be explained on the basis of a long central delay of inhibition for the following reason. Although the onset latency of the late positivity recorded in the experiment illustrated in Fig. 5 was 85 ms, this value was considerably shorter than that time interval (150 ms) between the end of the slow negative wave and the point closest to its beginning at which the shock applied to the paramedian nucleus became ineffective (Fig. 5E). These observations raise two important possibilities concerning the interactions between those central elements responsible for the generation and entrainment of the slow wave to the cardiac cycle. First, the
slow wave of SND probably results as the consequence of "avalanche excitation" transmitted through an interconnected population of brainstem neurons. This would account for the similar duration of each slow wave observed either before or after baroreceptor denervation (Fig. 3). Concerning this point, axon collaterals connecting large numbers of reticulospinal neurons have been described at medullary and pontine levels (5, 38, 39). Second, the ineffectiveness of stimuli applied to the paramedian nucleus soon after avalanche excitation was initiated suggests that the sympathoinhibitory effect of the baroreceptors was exerted most likely on interneuronal elements which trigger the neural network responsible for the slow wave. As a result, baroreceptor nervous discharge no longer would be effective in influencing the formation of the slow wave once the most rostral elements of the interconnected population of brainstem neurons were excited. In addition, triggering of the next wave of avalanche excitation would occur only after baroreceptor nervous discharge fell below some critical level. In this way, the slow wave of SND would become entrained to the cardiac cycle.

It can be assumed that the late positive potential reflects the capacity of the baroreceptor reflexes to entrain the slow wave to the cardiac cycle since it approximates the shape of the spontaneously occurring oscillation of SND. On this basis, it becomes further apparent that the sympathoinhibitory effect responsible for the entrainment occurred in the brainstem. The late positive wave produced by baroreceptor nerve or paramedian nucleus stimulation persisted after midcollicular transection. This observation indicates that a forebrain site or loop was not involved in the entrainment of the slow wave. Splanchnic nerve discharges evoked from descending spinal pressor tracts were not inhibited during the time course of the late positive potential. Thus, it is unlikely that sympathoinhibition represented by the late positivity occurred at a spinal site. By elimination, it can be concluded that sympathoinhibition of baroreceptor origin leading to the entrainment of the slow wave of SND to the cardiac cycle was mediated at a brainstem synapse.

The results of this study support previous reports from our laboratory (16, 40) concerning the existence of a spinal component of baroreceptor-induced sympathoinhibition. The pressor action of norepinephrine was associated with inhibition of splanchnic nerve discharges evoked by trains of pulses applied to descending pressor tracts in the midcervical spinal cord. The degree of depression of the splanchnic nerve responses was significantly greater than that produced by $C_1$ transection of the spinal cord. It was also demonstrated that the splanchnic nerve discharge evoked from descending spinal pressor tracts was depressed during the time course of the early positive potential elicited by stimuli applied to the paramedian nucleus. This observation suggests that the early positivity monitored sympathoinhibition exerted at a spinal level. Gootman and Cohen (18) reached the same conclusion since the onset latency of the early positivity evoked from the medullary depressor region was less than that for the splanchnic nerve discharges elicited from the medullary pressor region of the cat. That the early phase of positivity evoked by stimulation of the paramedian nucleus was of baroreceptor origin and monitored inhibition of transmission in vasopressor pathways was indicated by the following observations. First, trains of pulses applied to the carotid sinus or aortic depressor nerve produced an early positive potential on the splanchnic nerve. Second, the early positive potential could be monitored from postganglionic renal vasconstrictor fibers.

In previous reports from this laboratory (16, 40), potentials evoked in the external carotid postganglionic sympathetic nerve of the cat by trains of pulses applied to forebrain, brainstem, and spinal pressor sites were classified into two groups. The first group contained long-latency potentials which were inhibited by baroreceptor reflex activation or depressor region stimulation, while the second group contained shorter latency potentials which were not inhibited. In a later study, Taylor and Gebber (41) showed that individual preganglionic neurons of the upper thoracic spinal cord act as the final common pathway for those systems mediating the baroreceptor reflex-"sensitive" and reflex-"insensitive" responses. This led to the conclusion that vasconstrictor influences are distributed from the brain to the preganglionic neurons innervating the external carotid nerve over two distinct systems of pathways. However, in the present study, baroreceptor reflex-insensitive responses could not be demonstrated on the splanchnic nerve. Independent of onset latency, each and every splanchnic nerve response evoked from medullary and spinal pressor sites was inhibited by baroreceptor reflex activation. Thus, the central components of vasoconstrictor pathways distributed to the beds innervated by the splanchnic and external carotid nerves are organized quite differently.

In summary, this study has provided information concerning the functional organization of the baroreceptor reflex arc, as well as of those components of the central vasomotor system responsible for rhythmic discharges in sympathetic nerves. The proposed relationships between central vasomotor elements that are responsible for generation of the 3 cycle/s periodicity of SND and the baroreceptor reflex

**FIG. 13.** Wiring diagram depicting proposed relationships between central vasomotor elements responsible for generation of slow wave of SND and baroreceptor mechanisms which entrain slow wave to cardiac cycle. Unfilled circles: excitatory connections. Filled circles: inhibitory connections. B, baroreceptor afferent; D, driver neuron which initiates avalanche excitation; ERS, interconnected population of excitatory reticulospinal neurons; IRS, inhibitory brainstem interneuron; IKS, inhibitory spinal interneuron; ERS, postganglionic sympathetic neurons; T, tonic excitatory input.
arc are shown in Fig. 13. The wiring diagram admittedly is oversimplified and, in part, hypothetical. However, it is intended to provide a reasonable model for future research based on the results of the present investigation. The slow wave of SND is presumed to arise from avalanche excitation transmitted through an interconnected population of pontomedullary reticulospinal neurons (ERS). The reticulospinal nerve net is triggered at a frequency approximating 3 cycles/s by a driver neuron (D) which most likely receives tonic excitatory input (T) from unknown sources. Alternatively, the driver neuron could be a pacemaker. Brainstem sympathoinhibition (Ins) of baroreceptor origin (B) locks the synchronized burst of SND in a 1:1 relation to the cardiac cycle, thereby limiting the frequency at which the slow wave is generated. Brainstem inhibition probably is exerted either on the driver neuron or the most rostral elements of the reticulospinal net since the slow wave was terminated when stimuli were applied to the baroreceptor reflex arc just before but not immediately after its onset. The brainstem component of baroreceptor-induced sympathoinhibition is thought to be responsible for the late positive potential recorded from the splanchic or renal nerves. Finally, a reticulospinal pathway (IrS) mediating a second component of baroreceptor-induced sympathoinhibition (Is) is shown. The spinal component of baroreceptor inhibition may control the amplitude of the slow wave of SND and is responsible for the early positive potential recorded from peripheral sympathetic nerves.

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