

Regional and functional differences in the distribution of vestibulosympathetic reflexes

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Kerman, I. A., and B. J. Yates. Regional and functional differences in the distribution of vestibulosympathetic reflexes. *Am. J. Physiol.* 275 (Regulatory Integrative Comp. Physiol. 44): R824–R835, 1998.—Although considerable evidence suggests that the vestibular system regulates sympathetic outflow during movement and changes in posture, little is known about relative vestibular influences on activity of different sympathetic nerves and sympathetic efferents with different functions. In the present study, we demonstrated that electrical stimulation of the vestibular nerve in the cat elicited responses in sympathetic nerves innervating the head and abdominal viscera. This observation suggests that activity of sympathetic efferents innervating multiple body regions is affected by vestibular signals. These responses were attenuated by >80% when blood pressure was increased to >160 mmHg. Because raising blood pressure decreases the responsiveness of vasoconstrictor fibers, the simplest explanation for these data is that the vestibular system provides particularly strong inputs to components of the sympathetic nervous system that regulate peripheral vascular resistance. Furthermore, the relative magnitude of vestibulosympathetic reflexes was over four times larger in one sympathetic nerve composed mainly of vasoconstrictor efferents (renal nerve) than another nerve (external carotid nerve) containing similar types of fibers. Collectively, these data indicate that the vestibular system has selective influences on sympathetic outflow to particular tissues and body regions.

cardiovascular; blood pressure; orthostatic hypotension

ONE ROLE OF THE SYMPATHETIC nervous system is to maintain stable blood pressure under a number of conditions; to perform this function, brain stem neurons that regulate sympathetic outflow integrate a variety of sensory inputs (11). The vestibular system, which detects the body's position in space and the direction and velocity of movements, has been shown to influence sympathetic function (for review, see Refs. 23 and 24). Selective nose-up vestibular stimulation in decerebrate cats, produced by rotation of the head on a fixed trunk after denervations to remove visceral and nonlabyrinthine somatic inputs that could be produced by head movement, results in increased sympathetic nerve activity (29) and blood pressure (22). Furthermore, elimination of vestibular inputs through bilateral eighth nerve transection increases the susceptibility of anesthetized (5) or awake (10) cats to experience orthostatic hypotension during nose-up body rotations.

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In addition, physiological experiments have demonstrated that the rostral ventrolateral medulla, the region of the brain stem that is predominantly responsible for providing excitatory drive to sympathetic preganglionic neurons that regulate blood pressure (11), receives vestibular inputs (26, 30). Taken together, these data suggest that the vestibular system plays a role in modulating sympathetic outflow during postural changes that challenge the maintenance of stable blood pressure (i.e., assuming an upright posture in humans or a nose-up position in quadrupeds).

Previous studies have shown that the vestibular system influences the activity of several sympathetic nerves, including the inferior cardiac nerve, the greater splanchnic nerve, and the renal nerve (REN) (see Ref. 23 for review). However, these previous experiments did not determine the relative size of vestibulosympathetic reflexes in comparison with responses that include maximal sympathetic nerve activation. Thus it cannot be ascertained whether the vestibulosympathetic responses reported earlier represent a small or large change in sympathetic outflow to different organs. Furthermore, these studies did not determine whether stimulation of the vestibular system only modulated the firing of sympathetic efferents involved in cardiovascular control or whether sympathetic outflow to smooth muscle related to other functions (e.g., smooth muscle in the wall of gastrointestinal organs or bladder) was also affected by vestibular signals.

The present study had two aims. The first goal was to determine whether the relative magnitude of vestibulosympathetic responses was larger for some sympathetic nerves than for others. One factor that differs between sympathetic nerves is the content of fibers that innervate blood vessels. Thus the second aim was to test the hypothesis that the vestibular system has stronger effects on sympathetic efferents that regulate peripheral vascular resistance than sympathetic fibers with other functions (e.g., controlling motility in the gastrointestinal tract or bladder). Addressing these aims is important in validating our overall presumption that vestibulosympathetic reflexes serve mainly to offset orthostatic hypotension during changes in posture, as was proposed previously (23, 24). If this theory is correct, then vasoconstrictor fibers should be more powerfully influenced by vestibular stimulation than other types of sympathetic efferents. Furthermore, sympathetic fibers innervating caudally located arterioles would presumably be more responsive to vestibular stimulation than fibers innervating blood vessels in the head. Electrical stimulation of vestibular afferents was used in these experiments so that the effects of maximal activation of the vestibular system could be

determined. To test whether vasoconstrictor fibers are particularly sensitive to vestibular stimulation, we used the previous observations that responsiveness of vasoconstrictor sympathetic fibers is inversely related to blood pressure and that activation of baroreceptor afferents inhibits the excitability of these sympathetic efferents that presumably innervate vascular smooth muscle (1, 2, 8, 9, 12). Thus we predicted that the magnitude of vestibulosympathetic reflexes would be inversely proportional to blood pressure.

METHODS

All the procedures used in this study conformed with the "Guiding Principles in the Care and Use of Animals" of the American Physiological Society and were approved by the University of Pittsburgh's Animal Care and Use Committee.

General surgical procedures. Data were obtained from 29 adult cats of either sex. To determine whether the presence of anesthesia affected the distribution of vestibulosympathetic reflexes, we conducted experiments on both decerebrate animals and animals anesthetized using α -chloralose and urethan. Sixteen animals were initially anesthetized with 2–3% halothane (Fluothane, Ayerst Laboratories) vaporized in nitrous oxide and oxygen and after bilateral ligation of the common carotid arteries were rendered decerebrate via a midcollicular transection. After the decerebration was complete, anesthesia was removed. The other 13 animals were initially anesthetized with ketamine (15–20 mg/kg im) and xylazine (0.1–0.2 mg/kg im), an intravenous catheter was inserted, and anesthesia was maintained by injecting an α -chloralose-urethan mixture (initial dose of 40 mg/kg α -chloralose and 200 mg/kg urethan iv, supplemented every 2 h or as needed with an injection of 10% of the original dose). The depth of anesthesia was determined by monitoring blood pressure, respiratory rate, expression of withdrawal reflexes to pinch stimuli, and pupillary size. In all animals, the trachea was intubated and both femoral veins were cannulated for drug and fluid delivery. Blood pressure was monitored with the use of a Millar blood pressure transducer inserted 8–10 cm proximally through the femoral artery into the abdominal aorta. Core body temperature was measured and maintained between 36 and 38°C using a heating pad and heat lamp. After the completion of all surgical procedures and the determination of thresholds for vestibular nerve stimulation (see *Procedures for stimulation of vestibular nerve and determination of threshold current intensities for activating vestibular afferents*), paralysis was produced using gallamine triethiodide (Sigma, St. Louis, MO; initial injection of 10 mg/kg iv, maintained by hourly injections of 5 mg/kg), and artificial ventilation with room air (20 cycles/min) was begun. End-tidal CO₂ was monitored and maintained between 4 and 4.5% by adjusting tidal volume. In chloralose-urethan-anesthetized animals, paralysis was allowed to wear off every 3–4 h and the depth of anesthesia was confirmed by testing for withdrawal reflexes to pinch stimuli. Atropine (0.4 mg im) and dexamethasone (10–20 mg iv) were administered to all animals to reduce salivation and cerebral edema, respectively. At the end of the experiment, each animal was killed using pentobarbital sodium (120 mg/kg iv).

Procedures for stimulation of vestibular nerve and determination of threshold current intensities for activating vestibular afferents. The vestibular nerves were prepared for bipolar electrical stimulation with use of a previously described method (e.g., see Refs. 15, 16, 25, 27, and 28). On one or both sides, the tympanic bulla was exposed using a ventrolateral approach and was opened to expose the promontory. The

anterior wall of the promontory was opened to expose the scala vestibuli. One silver-silver chloride ball electrode, insulated except at the tip, was inserted into the scala vestibuli in the direction of the vestibule. The second electrode was placed 1–2 mm away, in the vicinity of the oval window. The effectiveness of vestibular nerve stimulation was determined by monitoring eye movements and neck contractions, which occur as part of vestibuloocular and vestibulocollic reflexes, respectively (21). These reflexes were elicited using a train of 50 shocks with a pulse width of 0.2 ms and an interpulse interval of 3 ms, repeated every 2 s. The positions of the electrodes were adjusted to produce a large differential between the stimulus intensity required to produce eye movements and that which resulted in facial movements. Contraction of facial muscles was most likely due to stimulus spread to motor fibers coursing in the facial nerve, which runs just outside the labyrinth and is the closest nontarget nerve to the stimulation site (7). The electrodes were then covered with warm semisolid paraffin and fixed in place with dental cement. In 15 experiments, a craniotomy and subsequent aspiration of the most caudal 5 mm of the midline cerebellum were performed so that a field potential elicited by vestibular nerve stimulation could be recorded from the medial longitudinal fasciculus (MLF). The MLF contains axons of second-order vestibular neurons, and the threshold of the MLF field potential elicited by stimulation of the vestibular nerve is only slightly higher than that of the field potential recordable from the vestibular nuclei (21). The MLF field potential was elicited using a single 0.2-ms shock and was recorded using a low-impedance (200 k Ω) glass-insulated tungsten electrode, amplified by a factor of 10⁵, and displayed on an oscilloscope.

Dissection of sympathetic nerves. After implantation of electrodes in the labyrinth, sympathetic nerves innervating different organs and body regions were dissected. Using a ventral approach, we exposed nerves innervating the head and neck [external carotid nerve (ECN)], the kidney (REN), the adrenal gland [adrenal nerve (ADN)], the gastrointestinal system [superior mesenteric nerve (SMN) and lumbar colonic nerve (LCN)], and the bladder and pelvic organs [hypogastric nerve (HGN)]. In one animal, recordings were also made from the inferior cardiac nerve, and responses were recorded from the celiac nerve in two animals. It was not possible to dissect every nerve in each animal, and thus a different combination of nerves was studied in each experiment.

In most of the experiments, a midline abdominal incision along the linea alba was performed. To facilitate access to abdominal and pelvic nerves, the superior and inferior mesenteric arteries and the portal vein were ligated, and the portion of the gut from the duodenum to the rectum was surgically removed. SMN filaments were identified as postganglionic nerves originating from the superior mesenteric ganglia and running along the superior mesenteric artery. REN branches were identified as the fine nerve filaments running along the renal artery and vein and entering the kidney at the renal pelvis. LCN and HGN were identified as postganglionic nerves originating from the inferior mesenteric ganglion and coursing either along the inferior mesenteric artery or toward the bladder, respectively. The celiac nerve was identified as postganglionic fibers arising from the celiac ganglion and running along the celiac artery. Thoracic splanchnic nerves were also dissected as they terminated in the celiac and the superior mesenteric ganglia, and lumbar splanchnic nerves were isolated as they terminated in the inferior mesenteric ganglia. Because the preganglionic nerves were dissected at the point where they entered the ganglion, it was possible to verify that all major inputs to the ganglion had been isolated.

In the experiments in which activity of ECN was recorded, a midline ventral incision in the neck was made and upper cervical portions of the trachea and the esophagus were removed. The right or left superior cervical ganglion and the cervical sympathetic trunk were gently separated from the vagus nerve and the nodose ganglion and dissected free from the surrounding connective tissue. ECN was identified as nerve filaments emanating from the superior cervical ganglion and coursing toward the external carotid artery. In experiments in which activity of ADN was recorded, the adrenal gland was approached through a left flank incision. ADN was identified as the fine filaments innervating the adrenal gland. The inferior cardiac nerve was isolated in one experiment near its origin from the stellate ganglion after removal of the heads of the first and second ribs.

Procedures for stimulating and recording from sympathetic nerves. Sympathetic nerves were covered with warm mineral oil or a mixture of warm mineral oil and petroleum jelly. Monopolar or bipolar recordings of nerve activity were made using low-resistance silver hook electrodes. Nerve activity was amplified (factor of 10^4 – 10^5), filtered with a band pass of 10–10,000 Hz, full-wave rectified, integrated (1-ms time constant), and digitized at a sampling rate of 1,000 Hz (using a 1401-Plus analog-to-digital converter manufactured by Cambridge Electronic Design). Digitized data were acquired by a Macintosh Quadra 800 computer and averaged online using the Spike-2 software package (Cambridge Electronic Design).

Nerves carrying preganglionic sympathetic fibers were stimulated using bipolar silver hook electrodes. Preganglionic nerves were stimulated to produce maximal activation of the postganglionic nerves so that the magnitude of vestibulosympathetic reflexes could be quantified (see *Experimental design*). In experiments in which the REN and SMN were studied, we stimulated all of the thoracic splanchnic nerves entering the superior mesenteric and celiac ganglia (which are fused with each other) together by laying the nerves across the leads of the same stimulating electrode. In experiments in which activity of the LCN and HGN were recorded, a similar procedure was used to simultaneously stimulate all of the lumbar splanchnic nerves entering the inferior mesenteric ganglion. Maximal activation of ECN was produced by stimulation of the cervical sympathetic trunk.

Experimental design. To elicit vestibulosympathetic reflexes, vestibular afferents were stimulated using five-shock trains (0.2-ms shock duration, 3-ms intershock interval) delivered every 1–2 s. A wide range of stimulation intensities (between 60 and 600 μ A) was used to determine maximal responses to vestibular stimulation. In animals in which both the ipsilateral and contralateral vestibular nerves were stimulated, responses elicited from each side were considered as separate cases. Vestibular afferents innervate hair cells in both semicircular canals and otolith organs that signal head movements in many different directions. Because the population of afferents activated depended on the placement of labyrinthine electrodes, it was likely that stimulation on the two sides excited populations of afferents signaling head movements in different directions and thus produced different patterns of responses. Nerves carrying preganglionic fibers (cervical sympathetic trunk, thoracic, and lumbar splanchnic nerves) were stimulated with single 0.2-ms-wide square-wave pulses delivered every 1–2 s and at intensities of 10–1,500 μ A.

Nerve responses were averaged online over several stimulus trials (10–40 for preganglionic stimulation, 80–300 for vestibular stimulation). Response sizes were determined by measuring the area under the response waveform (i.e., com-

bined area of positive and negative deflections from baseline), which was divided by baseline nerve activity. We standardized response area measurements to baseline activity to account for possible baseline fluctuations during the recording session. To enable comparisons of the size of vestibulosympathetic responses recorded from different nerves, we expressed maximal sympathetic nerve responses to vestibular stimulation as a percentage of the maximal nerve activation elicited by preganglionic stimulation.

To study the relationship between blood pressure levels and the expression of vestibulosympathetic reflexes, we delivered vestibular stimulation at normal resting levels of blood pressure and during periods of high blood pressure produced by intravenous infusions of the adrenergic agonist Aramine (metaraminol bitartrate; Merck Sharpe & Dohme, West Point, PA) or lactated Ringer solution. Alternatively, vestibular stimulation was triggered by the QRS complex of the electrocardiogram, and the delay was set so that the stimulus was delivered during systole or diastole or at intermediate blood pressure levels. Fluid or Aramine infusions were also used in some cases to increase systolic blood pressure levels during trials in which the stimulus was triggered from the electrocardiogram. Influences of blood pressure on the magnitude of vestibulosympathetic reflexes were only studied in animals anesthetized using α -chloralose-urethan, because ligation of the common carotid arteries in decerebrate animals (which was necessary to prevent bleeding from brain transection) may have altered the gain of the baroreceptor reflexes.

In some experiments, hexamethonium bromide (5–10 mg/kg iv) or tetraethylammonium (10–15 mg/kg iv) were administered to produce ganglionic blockade. This procedure allowed us to determine noise levels in the nerves recorded and to ascertain whether responses were exclusively due to changes in activity of postganglionic sympathetic fibers.

Controls for stimulus spread. To verify that the observed responses were due to stimulation of the vestibular nerve and not the result of current spread to nontarget afferents, in seven experiments we recorded vestibulosympathetic reflexes before and after inactivation of the medial and inferior vestibular nuclei, using lidocaine injections. These vestibular nuclei were targeted because previous studies have shown that portions of the medial and inferior vestibular nuclei located just caudal to the lateral vestibular nucleus mediate vestibulosympathetic reflexes (18, 28, 29). Lidocaine hydrochloride (2% by volume) saturated with fast green dye was injected into the medial and inferior vestibular nuclei after removal of the cerebellum. Six to ten injections (0.1 μ l each), spread \sim 0.5 mm apart, were made in two planes along the rostrocaudal extent of the vestibular nuclei on the same side as the stimulated vestibular nerve. In one additional experiment, the effects of aspiration of portions of the medial and inferior vestibular nuclei on the size of vestibulosympathetic reflexes were determined. At the end of every experiment, the brain stem was removed, postfixed in 10% Formalin, and cut into 100- μ m coronal sections and every other section was stained with thionine. The extent of dye spread or tissue damage was determined from camera lucida drawings of the sections.

Statistical analysis. Mean threshold differences in eliciting eye and facial movements in response to vestibular stimulation were evaluated using Student's *t*-test. Differences in the magnitude of vestibular stimulation-elicited reflexes recorded from different sympathetic nerves were evaluated using an analysis of covariance (ANCOVA) with recording site (different sympathetic nerves) as the factor and laterality of the vestibular stimulus, baseline nerve activity, and animal variability as covariates. Differences due to an animal's sex or

the type of preparation (chloralose-urethan anesthesia vs. decerebration) did not reach significance ($P < 0.05$, Student's t -test) and were not included in the model. A significant effect among nerves was evaluated post hoc with Student's t -tests. The relationships between blood pressure and sympathetic nerve activity and between blood pressure and the magnitude of vestibulosympathetic reflexes were evaluated using a least-squares linear regression and Pearson's correlation analysis. Similarly, we evaluated the relationship between level of blood pressure and amplitude of spontaneous fluctuations in sympathetic nerve activity using a linear regression analysis. Differences in the slopes of linear regression lines were evaluated using an ANCOVA model. Data are presented in the text as means \pm SE.

RESULTS

Stimulus intensities required to produce vestibulosympathetic reflexes and controls for stimulus spread. In 27 of the 29 animals (15 anesthetized using α -chloralose-urethan and 12 that would eventually be rendered decerebrate), the intensities of labyrinthine stimulation required for eliciting ipsilateral eye and facial movements were determined. Because a midcollicular decerebration can damage the third cranial nerve, we made threshold measurements before this procedure, while animals were anesthetized using halothane. Eye movement thresholds ranged from 50 to 480 μ A (mean of $143 \pm 11 \mu$ A). In contrast, the minimal intensities needed to generate facial movements ranged from 120 to $>1,000 \mu$ A (mean of $484 \pm 38 \mu$ A). These threshold differences were statistically significant ($P < 0.01$, paired Student's t -test). Choice of anesthesia (halothane or chloralose-urethan) did not affect the eye or facial movement thresholds in response to stimulation of the labyrinth. In 15 animals, MLF field potentials elicited by vestibular nerve stimulation were recorded. The minimal current intensities required to produce these responses ranged from 20 to 70 μ A (mean of $41 \pm 3 \mu$ A) and were always lower than the current intensities needed to elicit eye movements.

To verify that the observed sympathetic nerve responses were produced by activation of vestibular afferents and were not due to current spread from the labyrinthine electrodes to nontarget nerves, we compared vestibulosympathetic reflexes recorded before and after the vestibular nuclei were lesioned. During these control experiments, higher current intensities ($\sim 600 \mu$ A) were used than were typically used in other runs to elicit vestibulosympathetic reflexes. In four of seven animals, injections of lidocaine into the vestibular nuclei abolished vestibulosympathetic reflexes. Diffusion of lidocaine (estimated from spread of dye) in these animals was largely confined to the medial and inferior vestibular nuclei and the immediate surrounding areas of the brain stem. In an additional experiment, an aspiration lesion limited to the medial and inferior vestibular nuclei eliminated sympathetic nerve responses to vestibular nerve stimulation. However, in three of the experiments, injections of lidocaine were ineffective in attenuating sympathetic nerve responses to labyrinthine stimulation. In two of these cases, the

dye marking the injection sites was confined to small portions of the medial and inferior vestibular nuclei, and in the other animal the dye was located in portions of the inferior vestibular nucleus (with minimal spread to medial nucleus) while also diffusing caudally to the cuneate nucleus and ventrally to the spinal trigeminal nucleus and tract. These observations demonstrate that the inferior and medial vestibular nuclei must be functional for labyrinthine stimulation to produce large changes in sympathetic nerve activity. Thus it appears that the sympathetic nerve responses recorded in the present study were the result of stimulation of vestibular afferents.

Distribution of vestibulosympathetic reflexes and effects of ganglionic blockade on the responses. Sympathetic nerve responses to ipsilateral or contralateral vestibular stimulation were recorded from the ADN (5 cats), ECN (5 cats), REN (13 cats), SMN (10 cats), LCN (12 cats), and HGN (14 cats). In addition, we demonstrated that the inferior cardiac nerve responded to vestibular stimulation in one animal and we recorded vestibulosympathetic responses from the celiac nerve in two cases. Typically, recordings were made from several nerves in an animal, although the combination of nerves studied varied from experiment to experiment. Alterations in sympathetic nerve activity were elicited in all the nerves included in this study (see Fig. 1). Responses consisted of excitation, inhibition, or, in many cases, of a combination of excitation and inhibition. Similar complex waveforms were previously recorded from the splanchnic nerve after electrical vestibular stimulation (16, 28). Although the response pattern was variable from animal to animal, the response shapes were similar in all nerves recorded in a particular animal (see Fig. 1 for illustration). Thus, depending on the population of vestibular afferents preferentially activated by vestibular nerve stimulation in a particular experiment (i.e., the head direction encoded by the afferents that were excited), a similar change in activity was elicited across multiple sympathetic outflows.

In some animals, a variety of stimulus intensities was delivered so that the threshold for producing vestibulosympathetic reflexes could be determined. Mean intensities of vestibular nerve stimulation needed to elicit sympathetic nerve responses are listed in Table 1. This table also shows the onset latencies of sympathetic nerve responses to vestibular nerve stimulation. Responses to ipsilateral and contralateral vestibular stimulation had similar onset latencies and thresholds.

In 14 animals, noise levels were determined in some sympathetic nerves (ECN, REN, SMN, LCN, and HGN) postmortem or after systemic administration of the ganglionic blockers hexamethonium bromide or tetraethylammonium to abolish activity in sympathetic efferents. The levels of noise detected after death or administration of ganglionic blockers were indistinguishable, and average spontaneous nerve activity during the recording of vestibulosympathetic reflexes was 4.7 ± 0.5 ($n = 38$) times higher than noise levels. Because background activity was high, it was unlikely

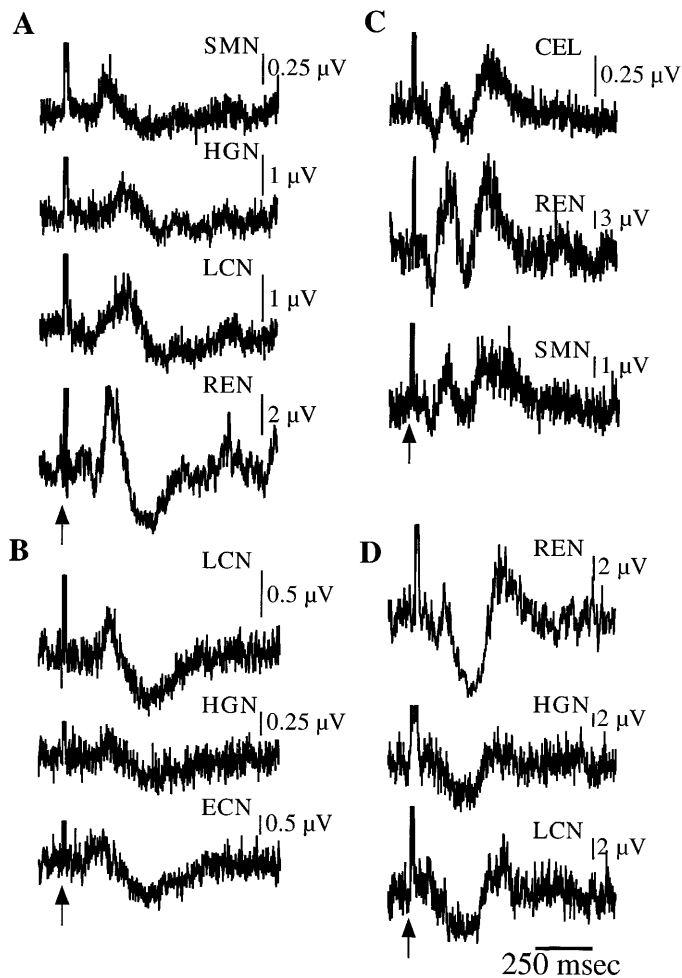


Fig. 1. Averaged sympathetic nerve responses to electrical stimulation of vestibular afferents. Each panel shows responses obtained from the same animal. Similar patterns of responses (excitation, inhibition, or combination of excitation and inhibition) were recorded from all nerves studied within the same animal. Arrows mark onset of 5-shock stimulus train. For some traces, large stimulus artifacts were truncated to conserve space. *A*: responses of superior mesenteric (SMN), hypogastric (HGN), lumbar colonic (LCN), and renal (REN) nerves to contralateral vestibular nerve stimulation at an intensity of 400 μ A. In this animal, all nerves exhibited a prominent excitation (upward deflection from baseline) that was followed by inhibition (downward depression). *B*: vestibulosympathetic responses recorded from LCN, HGN, and external carotid nerve (ECN) in another animal. These responses, consisting of excitation followed by a more prominent inhibition, were elicited by ipsilateral vestibular nerve stimulation at an intensity of 400 μ A. *C*: recordings from an animal in which ipsilateral vestibular stimulation (at an intensity of 500 μ A) produced biphasic excitatory responses in celiac nerve (Cel), REN, and SMN. *D*: recordings of predominantly inhibitory vestibulosympathetic reflexes elicited by stimulation of ipsilateral labyrinth at an intensity of 350 μ A.

that inhibitory nerve responses were not recorded or were underestimated. In two animals, we also compared sympathetic nerve responses to vestibular stimulation before and after systemic administration of hexamethonium. Ganglionic blockade completely abolished vestibular stimulation-elicited activity in the nerves that were studied, demonstrating that only postganglionic fibers mediated the observed changes in sympathetic nerve activity.

Regional differences in distribution of vestibulosympathetic reflexes. The magnitude of vestibulosympathetic reflexes was quantified in nine decerebrate and nine chloralose-urethan-anesthetized animals. Within each experiment, the area under the response waveform was measured for each nerve and standardized to the baseline nerve activity. To compare the magnitude of vestibular stimulation-evoked sympathetic nerve responses recorded from different nerves, the size of vestibulosympathetic reflexes was expressed as a fraction of maximal nerve activation produced by preganglionic nerve stimulation (see Fig. 2). Maximal response areas elicited by stimulation of the vestibular nerve or sympathetic preganglionic nerves were determined by comparing the size of responses to a variety of stimulus intensities. Stimulation of preganglionic nerves produced powerful, short-latency discharges in all of the postganglionic nerves (see Fig. 2 for examples). Because all visible nerves entering a sympathetic ganglion were stimulated, postganglionic nerve responses elicited by preganglionic stimulation presumably reflected activation of the large majority of postganglionic fibers. These responses were always abolished after hexamethonium bromide administration, verifying that they represented activity of postganglionic fibers. No difference ($P > 0.3$) in the size of vestibulosympathetic reflexes was noted between decerebrate and chloralose-urethan-anesthetized cats. For all sympathetic nerves studied, the magnitude of vestibular stimulation-elicited responses was $19 \pm 3\%$ ($n = 38$) of the maximal response to preganglionic nerve stimulation in decerebrate animals and $16 \pm 2\%$ ($n = 33$) in animals anesthetized with chloralose-urethan. Likewise, the laterality of vestibular stimulation had no apparent effect ($P > 0.3$) on nerve response sizes. Stimulation of the ipsilateral vestibular nerve evoked a response that was $19 \pm 3\%$ ($n = 30$) of the maximal response to preganglionic stimulation, whereas contralateral vestibular nerve stimulation elicited a response that was $16 \pm 2\%$ ($n = 41$) of the maximal sympathetic nerve activation produced by preganglionic stimulation. Therefore, data collected from decerebrate and anesthetized cats as well as responses elicited using ipsilateral and contralateral vestibular stimulation were pooled together for comparison. The magnitude of maximal vestibulosympathetic reflexes was compared with the size of maximal responses to preganglionic nerve stimulation for the following nerves: ECN (5 animals), REN (9 animals), SMN (8 animals), LCN (10 animals), and HGN (13 animals). As indicated in Fig. 3, vestibulosympathetic reflexes were 7–34% of the size of maximal postganglionic nerve responses to stimulation of preganglionic fibers. The relative sizes of vestibulosympathetic responses differed considerably from nerve to nerve; these differences were statistically significant ($P < 0.01$, ANCOVA). Post hoc statistical analysis (Student's *t*-test) revealed significant differences between the relative response magnitudes for the nerves studied (see Table 2). The size of vestibulosympathetic reflexes in REN (34% of response to preganglionic nerve stimulation) was greater than in all of the other

Table 1. Threshold current intensities required to elicit vestibulosympathetic reflexes and onset latencies of these responses

Nerve	Ipsilateral Vestibular Nerve				Contralateral Vestibular Nerve			
	Threshold, μA	<i>n</i>	Latency, ms	<i>n</i>	Threshold, μA	<i>n</i>	Latency, ms	<i>n</i>
ECN			69 \pm 15	3	153 \pm 73	3	84 \pm 14	5
REN	149 \pm 31	7	112 \pm 13	9	149 \pm 26	9	95 \pm 10	11
SMN	180 \pm 34	5	101 \pm 10	7	199 \pm 45	7	107 \pm 11	10
LCN	134 \pm 20	11	110 \pm 11	12	115 \pm 18	10	114 \pm 10	14
HGN	163 \pm 19	12	139 \pm 17	14	184 \pm 20	10	118 \pm 13	14
ADN					187 \pm 13	3	144 \pm 28	3

Values are means \pm SE; *n* = number of observations. Onset latencies and thresholds were determined for responses elicited by a train of 5 shocks; onset latencies were measured from first shock in stimulus train. Only 2 cases were available where values are missing, and thus an average is not provided. ECN, external carotid nerve; REN, renal nerve; SMN, superior mesenteric nerve; LCN, lumbar colonic nerve; HGN, hypogastric nerve; ADN, adrenal nerve.

nerves studied ($P < 0.05$). In contrast, vestibulosympathetic reflexes in ECN (7% of response to preganglionic nerve stimulation) were smaller than in all the other nerves ($P < 0.05$) except HGN ($P = 0.15$).

Relationship between blood pressure and expression of vestibulosympathetic reflexes. Because excitability of sympathetic efferents controlling peripheral vascular resistance (i.e., vasoconstrictor fibers), but not sympathetic efferents with other functions such as regulating motility, is presumably altered by changes in blood pressure (1, 2, 8, 9, 12), we compared the amplitude of vestibulosympathetic reflexes elicited at different blood pressure levels. Recordings were made from the following nerves during this analysis: ECN, REN, LCN, SMN, and HGN. These experiments were performed on five animals anesthetized using chloralose-urethan, in which the common carotid arteries were not ligated. In two animals, both the ipsilateral and contralateral vestibular nerves were stimulated; the responses to

stimulation on each side were treated as separate cases for this analysis. In five of the experiments, stimuli were delivered to the vestibular nerve randomly with respect to the cardiac cycle, while mean blood pressure was increased through intravenous infusions of Ringer lactate solution or the adrenergic agonist Aramine. Vestibulosympathetic reflexes were recorded from the following nerves during these trials: REN (3 cases), SMN (3 cases), LCN (1 case), and HGN (3 cases). As illustrated in Fig. 4, an increase in blood pressure resulted in a powerful attenuation in the magnitude of vestibulosympathetic reflexes.

In all experiments, vestibular nerve stimulation was additionally triggered by the R wave of the electrocardiogram in some runs and the delay between the trigger and the stimulus was varied to elicit responses at different parts of the cardiovascular cycle. In all but two experiments, Aramine was infused during some trials in which the stimulus was delivered at systole to further increase systolic blood pressure. Responses were recorded from the following nerves while vestibular stimuli were delivered during systole or diastole: ECN (3 cases), REN (3 cases), SMN (5 cases), LCN (3 cases), and HGN (4 cases). Figure 5 illustrates vestibulosympathetic reflexes elicited in four nerves during diastole and systole. As with trials in which the stimulus was delivered randomly with respect to the cardiac cycle and blood pressure was altered using infusions of

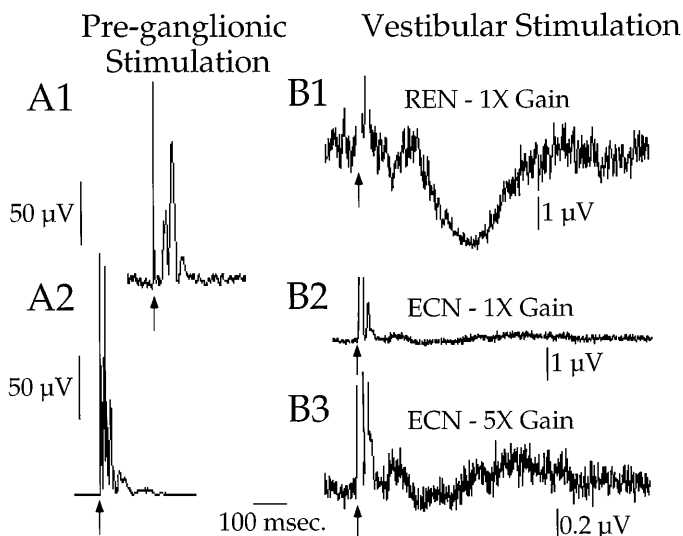


Fig. 2. Magnitude of vestibulosympathetic reflexes with respect to maximal activation of postganglionic nerves produced by stimulation of preganglionic fibers. Responses recorded from REN and ECN are shown; all traces were recorded from the same animal. Note that responses to preganglionic nerve stimulation (A) are shown at a much lower gain than those to vestibular stimulation (B) and that vestibulosympathetic responses recorded from REN are larger than those recorded from ECN.

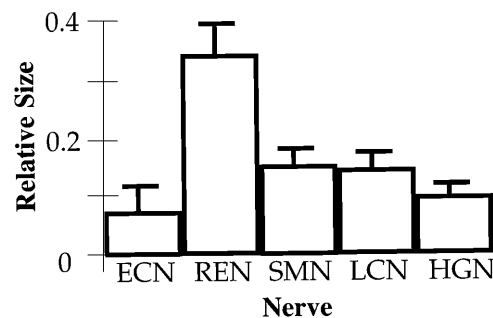


Fig. 3. Comparison of relative size of maximal vestibulosympathetic reflexes (as a fraction of maximal responses elicited by stimulation of preganglionic fibers) recorded in different sympathetic nerves. Analysis of covariance revealed statistically significant ($P < 0.01$) differences in relative sizes of responses; results of post hoc analysis are presented in Table 2. Error bars indicate SE.

Table 2. Comparisons of sizes of vestibulosympathetic reflex elicited in different nerves

	REN	SMN	LCN	HGN
ECN	*	*	*	0.152
REN		*	*	*
SMN			0.214	*
LCN				0.149

After initial analysis using analysis of covariance, between-nerve comparisons were made using Student's *t*-test. *Comparisons that produced statistically significant results ($P < 0.05$). Nonsignificant *P* values are given.

Aramine or Ringer lactate solution, vestibulosympathetic reflexes appeared to be much smaller when blood pressure was high than when it was low. In most cases, delivery of vestibular stimulation during systole produced no appreciable responses (see Fig. 5). In trials in which responses were not completely eliminated by stimulation during systole, further increasing systolic blood pressure levels with fluid or Aramine infusions abolished the vestibulosympathetic reflex.

However, it is possible that in some cases when stimuli were triggered from the electrocardiogram, responses were obscured by strong entrainment of baseline sympathetic activity to blood pressure fluctuations during the cardiac cycle (such entrainment is obvious in Fig. 5, *A* and *B*). To quantify data collected under different blood pressure conditions, it was necessary to eliminate entrainment of sympathetic nerve activity to blood pressure oscillations from the records. To do so, nerve activity in response to blood pressure fluctuations in the absence of vestibular stimulation was averaged and subtracted from records in which vestibular stimuli were presented, as illustrated in Fig. 6.

To quantify the effects of blood pressure on the magnitude of vestibulosympathetic reflexes, we plotted the peak-to-trough response amplitude (expressed as a percentage of the largest response recorded from a nerve) as a function of blood pressure. Pooled data from all experiments are shown in Fig. 7, which includes responses to vestibular nerve stimulation delivered at systole and diastole as well as responses to stimuli randomly delivered with respect to the cardiac cycle but at different mean blood pressure levels. In the former

case, the *abscissa* indicates the diastolic or systolic blood pressure at the time of stimulus presentation, whereas in the latter case the *abscissa* shows mean blood pressure during stimulus delivery (Fig. 7). As discussed in *Experimental design*, it was necessary to subtract spontaneous oscillations in nerve activity (that were presumably elicited by baroreceptor stimulation during the cardiac cycle) from vestibular system-evoked responses. This method may have resulted in a small error in the calculation of the magnitude of vestibulosympathetic reflexes. Nonetheless, a linear regression analysis revealed statistically significant ($P < 0.05$) correlations between response amplitude and blood pressure for ECN, REN, LCN, SMN, and HGN. For all nerves, increasing blood pressure to >160 mmHg attenuated the amplitude of vestibulosympathetic responses to $<20\%$ of the magnitude of responses elicited when stimuli were presented while blood pressure was 100 mmHg.

Comparison of relative amplitude of responses to vestibular nerve stimulation in sympathetic nerves presumed to be mainly composed of vasoconstrictor efferents. This study also sought to determine whether the relative strength of vestibular influences on vasoconstrictor efferents innervating rostral vascular beds differs from that on efferents innervating caudal vasculature. Previous experiments (4, 6, 12, 14, 19, 20) have suggested that two of the nerves included in this study, REN and ECN, are composed almost entirely of vasoconstrictor efferents. Because these two nerves innervate vasculature in different parts of the body (REN innervates renal blood vessels and ECN innervates vasculature of head and neck), a comparison of the relative size of vestibulosympathetic responses in REN and ECN may be useful to test the hypothesis that vasoconstrictor fibers innervating some vascular beds are more sensitive to vestibular inputs than others.

To confirm previous findings that ECN and REN are composed mainly of vasoconstrictor fibers, the entrainment of spontaneous discharges to blood pressure oscillations was quantified by relating systolic, diastolic, and mean blood pressure levels to synchronized changes in nerve activity. These measurements were made at resting and elevated blood pressure produced

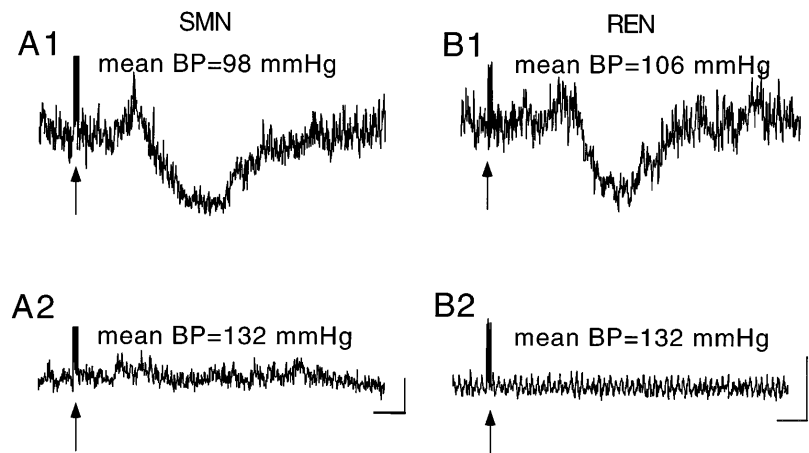


Fig. 4. Effect of increasing mean blood pressure (BP) on averaged vestibulosympathetic responses. Activity of SMN (*A*) and REN (*B*) was recorded at resting BP levels (*A1* and *B1*) and during a period of elevated BP (*A2* and *B2*) produced by Aramine infusion. Note that when BP was high, vestibulosympathetic responses were either weak or absent. Vestibular stimuli were delivered randomly with respect to cardiovascular cycle. Same stimulus intensity was used to elicit responses at both high and low BP. Vertical calibration, $1 \mu\text{V}$; time scale, 100 ms.

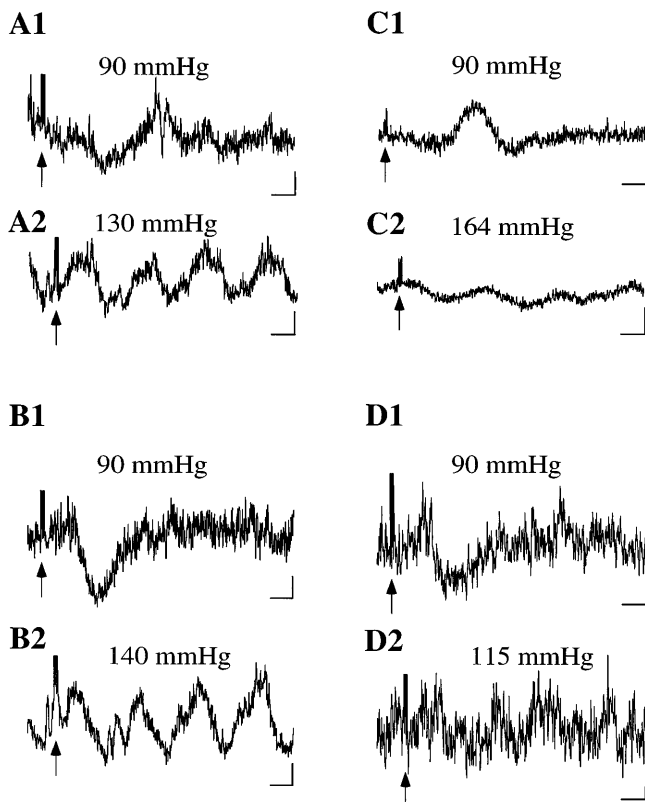


Fig. 5. Averaged vestibulosympathetic responses recorded from the REN (A), SMN (B), LCN (C), and HGN (D) nerves. Vestibular stimuli were either delivered during diastole (traces A1, B1, C1, and D1) or systole (traces A2, B2, C2, and D2). Intravenous Aramine infusions were used to increase systolic BP in traces A2, B2, and C2, but not in D2. Note that responses recorded during diastole are much larger than those recorded during systole. Records obtained when BP was increased using Aramine (traces A2, B2, and C2) also exhibit considerable entrainment of nerve activity to cardiac cycle. Data in A, B, and D were obtained from the same animal. Vertical scale bars represent 1 μ V in A, B, and D and 0.3 μ V in C; time scale bar indicates 100 ms.

by fluid or Aramine infusions. Multiple sympathetic nerves (ECN, REN, SMN, LCN, and HGN) whose activity was recorded in 13 chloralose-urethan-anesthetized cats were included in the analysis. Results were expressed as a fraction of a control measurement of nerve activity taken at the lowest blood pressure level. In each animal, the strength of correlation between spontaneous activity of a nerve and the level of blood pressure was determined from the slope of a least-squares regression line fit to the measurements made from each nerve. Spontaneous activity of ECN was strongly correlated with blood pressure, so that higher blood pressure levels strongly inhibited spontaneous nerve firing in five of five animals. A linear regression analysis of the pooled data revealed a statistically significant correlation between blood pressure and nerve activity ($r^2 = 0.89$, $P < 0.01$; Fig. 8A). The slope of the linear regression line was -1.43 . Activity of REN was similarly sensitive to blood pressure changes; in nine of nine animals, spontaneous nerve activity decreased at higher blood pressure levels, as demonstrated by a linear regression analysis of the pooled

data ($r^2 = 0.73$, $P < 0.01$; Fig. 8B). The slope of the linear regression line was -0.78 . These observations suggest that both ECN and REN contain a large number of vasoconstrictor fibers.

However, for this analysis to be meaningful, it was also necessary to demonstrate that sympathetic nerves containing a large number of fibers that do not innervate vascular smooth muscle have activity that is less sensitive to changes in blood pressure. For example, both LCN and HGN contain many motility-regulating fibers whose activity is unaffected by changes in blood

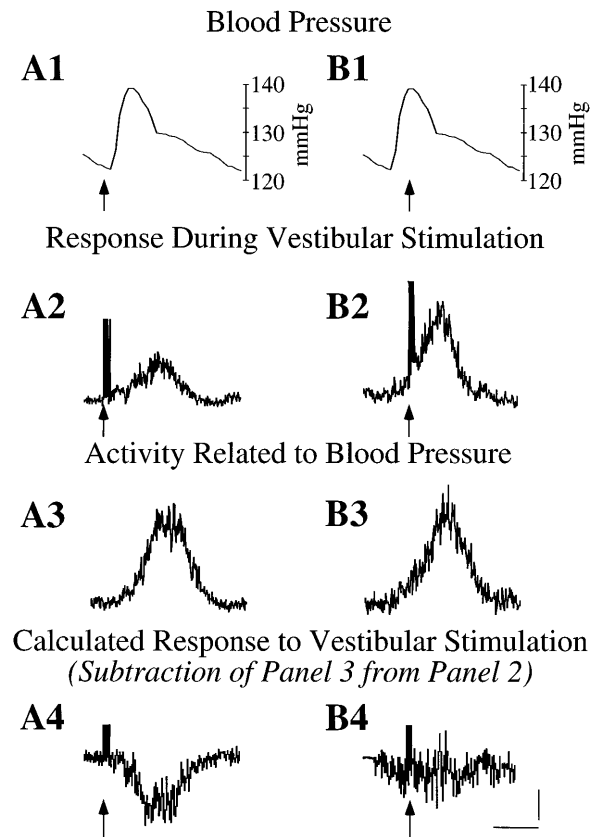
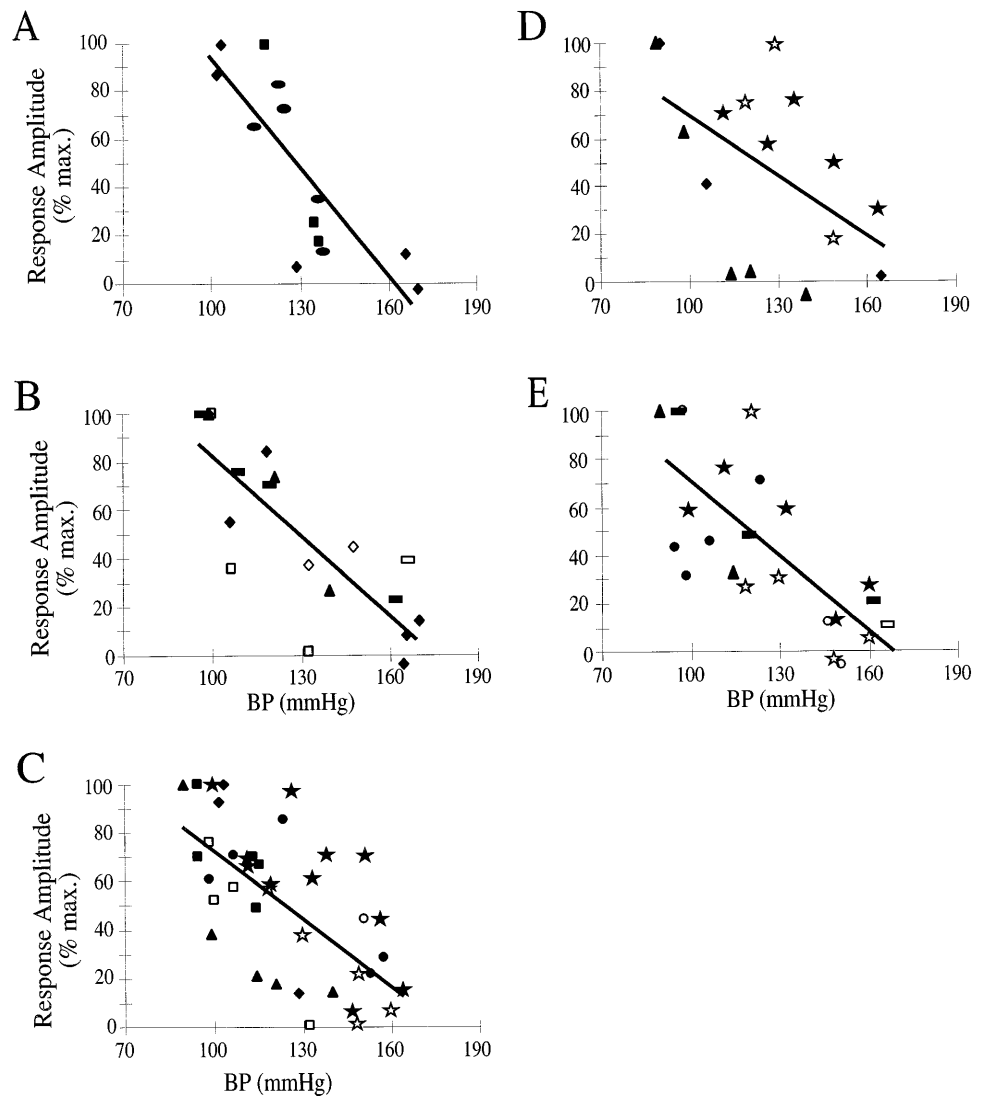


Fig. 6. Example of subtraction of entrained spontaneous nerve activity from vestibulosympathetic reflexes. Responses of ECN to contralateral vestibular stimulation (at an intensity of 500 μ A) delivered during diastole (BP of 123 mmHg, A) and systole (BP of 136 mmHg, B) were recorded. Top trace of each column shows arterial BP; arrow indicates onset latency of stimulus train. Traces A2 and B2 illustrate responses to vestibular nerve stimulation; because stimulation in each sweep was locked to cardiovascular cycle, considerable entrainment of baseline nerve activity to spontaneous BP fluctuations was superimposed on vestibulosympathetic responses. For elimination of this activity that was presumably related to activation of baroreceptor afferents, it was necessary to record sympathetic nerve activity that was synchronized with BP oscillations in absence of vestibular nerve stimulation (traces A3 and B3). These cardiac cycle-related oscillations in nerve activity were recorded during the same runs in which vestibular stimuli were presented, but at sufficiently long latency after stimuli were delivered to allow vestibulosympathetic responses to dissipate. Changes in nerve activity related to fluctuations in BP were averaged and subtracted from traces during which vestibular stimuli were delivered, so that responses elicited only by vestibular stimuli could be calculated (traces A4 and B4). This analysis revealed the presence of clear responses to vestibular stimulation during diastole that were abolished during systole. Vertical calibration bars indicate 1 μ V; time scale bars indicate 100 ms.

Fig. 7. Correlation between amplitude of vestibul sympathetic reflexes and BP levels is illustrated. Response amplitudes were expressed as a percent of the largest response recorded from a particular nerve. Different symbols demarcate measurements obtained in different cases. Filled symbols represent data obtained using electrocardiogram-triggered stimulation technique; open symbols indicate responses elicited by randomly delivered vestibular stimuli at resting and elevated BP. Note that in some experiments, both of these methods were used (explaining why open and closed symbols of same type appear in some panels). Correlation between magnitude of pooled responses from all animals and BP levels was examined using a linear regression analysis. Results of Pearson's correlations demonstrated that this relationship was statistically significant ($P < 0.05$) for all nerves: ECN (A) $r^2 = 0.70$, $n = 3$; REN (B) $r^2 = 0.67$, $n = 4$; LCN (D) $r^2 = 0.44$, $n = 5$; SMN (C) $r^2 = 0.29$, $n = 3$; and HGN (E) $r^2 = 0.56$, $n = 4$.



pressure (1, 4, 9, 19). As would be expected, the activity of both of these nerves was not as sensitive to changes in blood pressure as that of REN or ECN. A linear regression of pooled data collected from 10 animals revealed no statistically significant correlation between blood pressure and LCN activity ($r^2 = 0.22$, $P = 0.11$; Fig. 8D). In the case of HGN, although a linear regression analysis of pooled data from 13 animals revealed a statistically significant ($r^2 = 0.14$, $P < 0.01$) correlation between blood pressure and nerve activity, the slope of the regression line (-0.38) was not steep (see Fig. 8E). Figure 8F directly compares the slopes of all linear regression lines comparing nerve activity and blood pressure and illustrates that the slopes were steepest for REN and ECN and flattest for HGN and LCN.

To statistically evaluate the difference in the relationship between blood pressure and spontaneous nerve activity, ECN and REN were grouped together as the nerves that presumably had the highest content of vasomotor fibers, and LCN and HGN were grouped together as the nerves with the lowest content of

vasomotor fibers. An ANCOVA was performed with blood pressure as the continuous independent variable, vasoconstrictor fiber content (high, ECN and REN; low, HGN and LCN) as a factor, and nerve activity as a dependent variable. This analysis showed that the dependence between nerve activity and blood pressure was significantly different ($P < 0.05$) between the two groups, and therefore it confirms previous reports that ECN and REN have a higher content of vasoconstrictor fibers than LCN or HGN.

In contrast, another nerve included in this study (SMN) appeared to contain a large number of both vasoconstrictor fibers and motility-regulating efferents. An increase in blood pressure strongly attenuated activity in this nerve (see Fig. 8C); a linear regression analysis of pooled data collected from six animals revealed a significant correlation between blood pressure levels and SMN activity ($r^2 = 0.69$, $P < 0.01$). The slope of the linear regression line was -0.70 . This finding supports the previous observation that SMN, unlike LCN, has considerable activity that is synchronized with the cardiac cycle (4, 13). However, SMN

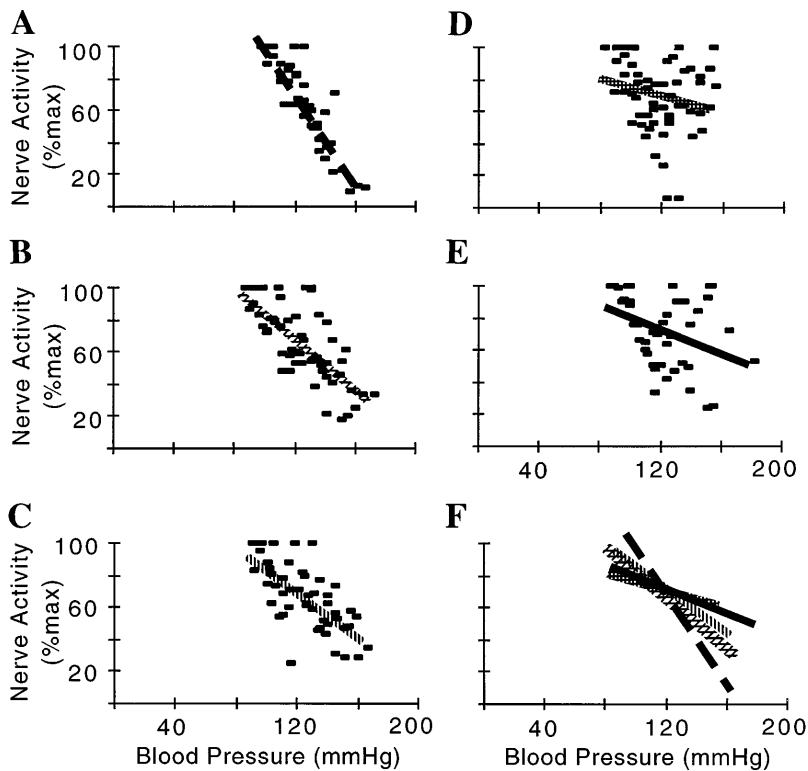


Fig. 8. Comparison of sensitivity of sympathetic nerves to changes in BP levels. Baseline entrainment of sympathetic nerve activity was quantified in the ECN (A), REN (B), SMN (C), LCN (D), and HGN (E) nerves. Measurements were made at resting BP and at elevated BP levels after fluid or Aramine infusions (see METHODS). Results were expressed as a fraction of nerve activity measured at lowest BP level in each experiment. Least-squares linear regression analysis was performed on data from each nerve. Linear regression lines for each nerve were plotted together (F) so that slopes could be directly compared. Nerve represented by each line type is indicated in A–E. Note that slopes of linear regression lines are steeper for REN and ECN than for other nerves, particularly HGN and LCN.

innervates the gastrointestinal system, and a previous study showed that a large fraction of fibers in this nerve lack cardiac cycle-related activity (20). Thus, despite the relationship between SMN activity and blood pressure observed in these experiments, this nerve must contain a number of motility-regulating fibers. Because SMN is a mixed nerve composed of a large number of both motility-regulating fibers and vasoconstrictor efferents, it was not included in the quantitative comparison between blood pressure levels and fluctuations in sympathetic nerve activity.

As discussed and as illustrated in Fig. 3, the relative size of vestibulosympathetic responses was significantly smaller in ECN than in REN, although both nerves appear to be mainly composed of vasoconstrictor fibers (see Fig. 8). Collectively, these observations suggest that vasoconstrictor efferents in ECN are influenced less strongly by vestibular signals than those in REN.

DISCUSSION

Electrical stimulation was used in the present experiments to activate vestibular afferents. One advantage of this mode of stimulation is that it can be used to activate all vestibular afferents simultaneously (21), which allows an examination of the effects of maximal activation of the vestibular system. Natural vestibular stimulation, on the other hand, is inherently weaker in that it produces submaximal excitation of only a subset of vestibular afferents (21). However, because electrical vestibular stimulation simultaneously activates all vestibular afferents, it provides a nonphysiological message to the central nervous system indicating that the

head is simultaneously moving in all possible directions. The presence of this complicated input explains why the vestibulosympathetic responses recorded in these experiments were composed of a complex combination of excitation and inhibition. Nonetheless, it was unlikely that stimulus spread to nonvestibular afferents contributed to the observed responses. Vestibulosympathetic responses could be produced by current intensities that were lower than those required to activate motor axons in the facial nerve, which runs just outside the labyrinth. Furthermore, vestibulosympathetic reflexes were strongly attenuated after inactivation of the medial and inferior vestibular nuclei with lidocaine injections or destruction of these vestibular nuclei using aspiration. The extent of reflex attenuation depended on the size of each lesion, so that the most effective lesions affected the greatest proportion of the medial and inferior vestibular nuclei. In combination, these observations suggest that the sympathetic nerve responses to vestibular nerve stimulation recorded in this study were the result of activation of vestibular afferents. Vestibulosympathetic reflexes could be recorded from all of the sampled sympathetic nerves, suggesting that vestibular influences are distributed to a wide variety of sympathetic outflows. However, the relative amplitude of the responses expressed as a fraction of maximal nerve excitation produced by stimulation of preganglionic efferents varied from nerve to nerve. The largest vestibulosympathetic reflex was recorded from REN (34% of area of maximal response to preganglionic nerve stimulation), and the smallest response was recorded from ECN (7% of area of maximal response to preganglionic nerve stimulation). This

observation suggests that the vestibular system does not have uniform influences on all sympathetic efferents.

Another finding was that raising blood pressure attenuated the size of vestibulosympathetic responses. This effect was obvious both when we compared responses elicited by stimulation during diastole and systole and when vestibular stimuli were delivered randomly with respect to the cardiac cycle but mean blood pressure was increased by infusing fluid or Aramine. For example, if blood pressure was increased to >160 mmHg, these responses were on average $<20\%$ of the magnitude of responses elicited when blood pressure was 100 mmHg. The simplest explanation for this finding is that activation of baroreceptor afferents inhibits neurons in the neural pathway that relays vestibular signals to sympathetic preganglionic neurons. Several areas of the brain stem that regulate sympathetic outflow receive vestibular signals, including the nucleus of the solitary tract (3, 27), the caudal ventrolateral medulla (15, 16, 17), the lateral tegmental field (17, 25), and the rostral ventrolateral medulla (26, 30). However, the only neurons shown to receive convergent baroreceptor and vestibular signals are located in the rostral ventrolateral medulla (26, 30). Neurons in the nucleus of the solitary tract and the caudal ventrolateral medulla that are part of the baroreceptor reflex arc do not respond appreciably to stimulation of the vestibular nerve (15, 27). Because neurons in the rostral ventrolateral medulla are powerfully inhibited by baroreceptor stimulation (11) and are essential for relaying vestibular signals to sympathetic preganglionic neurons (see Ref. 24 for review of literature), raising blood pressure should diminish the transmission of vestibular signals to the spinal cord by these cells.

Previous studies have shown that increasing blood pressure predominantly decreases the sensitivity of sympathetic vasoconstrictor fibers and not sympathetic efferents with other functions (e.g., those that regulate motility) (1, 2, 8, 9, 12). It follows from these observations that baroreceptor stimulation preferentially decreases the excitability of central nervous system pathways that regulate sympathetic outflow to vascular smooth muscle but does not appreciably influence pathways that control sympathetic outflow to other targets. Thus our findings suggest that vestibular signals have the strongest influences on components of the sympathetic nervous system that regulate peripheral vascular resistance.

However, it cannot be ruled out that some types of sympathetic fibers other than vasoconstrictor efferents are at least weakly influenced by the vestibular system. For example, some sympathetic efferents are silent in the types of preparations used in this study, and the excitability of these fibers may have been too low for vestibular stimulation to have attenuated their activity (8). Furthermore, the sympathetic nerves that were studied did not include all types of sympathetic fibers (e.g., sympathetic efferents mediating piloerection were not present in sampled nerves), limiting the conclu-

sions that can be made. Nonetheless, motility-regulating efferents in at least LCN and HGN are known to have spontaneous activity in the types of preparations used in this study (1, 8, 9), and it is unlikely that strong vestibular influences on these fibers would have been missed in our experimental paradigm.

These data also allowed us to test the hypothesis that the relative strength of vestibular influences on vasoconstrictor efferents innervating rostral vascular beds differs from that on sympathetic fibers innervating caudal vasculature. Blood pressure-related oscillations were larger in ECN and REN than in the other nerves recorded in these experiments, suggesting that these two nerves contain the highest fraction of vasomotor fibers. Our findings are in agreement with those of deGroat and Krier (4), who demonstrated that activity of the REN is much more sensitive to blood pressure fluctuations than that of the LCN. Single-fiber studies have confirmed that REN is composed of functionally uniform efferents with activity related to blood pressure (6, 20). Our finding that ECN activity is strongly related to the cardiac cycle is also not surprising, because this nerve is believed mainly to innervate the external carotid artery (14) and has previously been described as being composed mostly of vasoconstrictor fibers (19). Because REN and ECN are involved mainly in controlling vasoconstriction and innervate different regions of the body, we compared the magnitude of vestibulosympathetic reflexes recorded from these two nerves to determine whether caudal vasoconstrictor efferents are more powerfully influenced by the vestibular system than are rostral efferents. The relative size of the vestibular system-elicited response in ECN was $\sim 25\%$ of that in REN. One interpretation of this finding is that caudally located vasoconstrictor fibers are more sensitive to vestibular inputs than the rostral vasoconstrictor fibers and act to produce greater vasoconstriction in the caudal vascular beds to counteract development of orthostatic hypotension. Alternatively, it is possible that the activity of renal efferents is more sensitive to vestibular stimuli than other types of sympathetic fibers, perhaps to insure that the kidney always receives an adequate blood supply to maintain renal function. Thus further experiments that measure changes in blood flow in response to vestibular stimuli will be required to determine whether peripheral vascular resistance in the caudal body is more affected by labyrinthine inputs than that in the upper body. Additionally, recordings from single functionally identified sympathetic fibers will be needed to establish whether the vestibular system provides selective inputs to vasoconstrictor efferents innervating particular vascular beds.

Perspectives

This study demonstrated that although the vestibular system influences the activity of many sympathetic nerves, the effects are not uniform on all sympathetic outflows. In particular, components of the sympathetic nervous system that regulate vascular tone appear to be more sensitive to vestibular stimulation than sympa-

thetic efferents that regulate motility in the gastrointestinal system or the bladder. Furthermore, differences also appear to exist in the strength of vestibular influences on the activity of vasoconstrictor fibers innervating different vascular beds. Thus vestibular signals clearly do not elicit nonspecific "fight-or-flight" responses that involve simultaneous activation or inhibition of all components of the sympathetic nervous system, indicating that vestibular influences on the sympathetic nervous system serve a particular function. These findings support the hypothesis that vestibulosympathetic reflexes act selectively in correcting blood pressure during movement and changes in posture.

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REFERENCES

- Bahr, R., B. Bartel, H. Blumberg, and W. Jänig. Functional characterization of preganglionic neurons projecting in the lumbar splanchnic nerves: neurons regulating motility. *J. Auton. Nerv. Syst.* 15: 109-130, 1986.
- Bahr, R., B. Bartel, H. Blumberg, and W. Jänig. Functional characterization of preganglionic neurons projecting in the lumbar splanchnic nerves: vasoconstrictor neurons. *J. Auton. Nerv. Syst.* 15: 131-140, 1986.
- Balaban, C. D., and G. Beryozkin. Vestibular nucleus projections to nucleus tractus solitarius and the dorsal motor nucleus of the vagus nerve: potential substrates for vestibulo-autonomic interactions. *Exp. Brain Res.* 98: 200-212, 1994.
- DeGroat, W. C., and J. Krier. The central control of the lumbar sympathetic pathway to the large intestine of the cat. *J. Physiol. (Lond.)* 289: 449-468, 1979.
- Doba, N., and D. J. Reiss. Role of the cerebellum and vestibular apparatus in regulation of orthostatic reflexes in the cat. *Circ. Res.* 34: 9-18, 1974.
- Dorward, P. K., S. L. Burke, W. Jänig, and J. Cassell. Reflex responses to baroreceptor, chemoreceptor and nociceptor inputs in single renal sympathetic neurones in the rabbit and the effects of anaesthesia on them. *J. Auton. Nerv. Syst.* 18: 39-54, 1987.
- Gacek, R. R. The course and central termination of first order neurons supplying vestibular end organs in the cat. *Acta Otolaryngol. Suppl. (Stockh.)* 254: 1-66, 1969.
- Jänig, W., and E. M. McLachlan. Characteristics of function-specific pathways in the sympathetic nervous system. *Trends Neurosci.* 15: 475-481, 1992.
- Jänig, W., M. Schmidt, A. Schnitzler, and U. Wesselmann. Differentiation of sympathetic neurones projecting in the hypogastric nerves in terms of their discharge patterns in cats. *J. Physiol. (Lond.)* 437: 157-179, 1991.
- Jian, B. J., L. A. Cotter, K. R. Patterson, and B. J. Yates. Removal of vestibular inputs produces reduced orthostatic tolerance in awake cats. *Soc. Neurosci. Abstr.* 23: 723, 1997.
- Loewy, A. D., and K. M. Spyer. *Central Regulation of Autonomic Functions*. New York: Oxford University, 1990.
- Meckler, R. L., and L. C. Weaver. Characteristics of ongoing and reflex discharge of single splenic and renal sympathetic postganglionic fibres in cats. *J. Physiol. (Lond.)* 396: 139-153, 1988.
- Ninomiya, I., and H. Irisawa. Non-uniformity of the sympathetic nerve activity in response to baroreceptor inputs. *Brain Res.* 87: 313-322, 1975.
- Skok, V. I. *Physiology of Autonomic Ganglia*. Tokyo, Japan: Igaku Shoin, 1973.
- Steinbacher, B. C., and B. J. Yates. Processing of vestibular and other inputs by the caudal ventrolateral medullary reticular formation. *Am. J. Physiol.* 271 (*Regulatory Integrative Comp. Physiol.* 40): R1070-R1077, 1996.
- Steinbacher, B. C., and B. J. Yates. Brainstem interneurons necessary for vestibular influences on sympathetic outflow. *Brain Res.* 720: 204-210, 1996.
- Stocker, S. D., B. C. Steinbacher, C. D. Balaban, and B. J. Yates. Connections of the caudal ventrolateral medullary reticular formation in the cat brainstem. *Exp. Brain Res.* 116: 270-282, 1997.
- Uchino, Y., N. Kudo, K. Tsuda, and Y. Iwamura. Vestibular inhibition of sympathetic nerve activities. *Brain Res.* 22: 195-206, 1970.
- Weaver, L. C. Organization of sympathetic responses to distension of urinary bladder. *Am. J. Physiol.* 248 (*Regulatory Integrative Comp. Physiol.* 17): R236-R240, 1985.
- Weaver, L. C., R. L. Meckler, and R. D. Stein. Organization of sympathetic influences on the kidney and capacitance circulation. In: *Organization of the Autonomic Nervous System: Central and Peripheral Mechanisms*, edited by J. Ciriello, F. R. Calaresu, L. P. Renaud and C. Polosa. New York: Liss, 1987, p. 101-109.
- Wilson, V. J., and G. Melvill Jones. *Mammalian Vestibular Physiology*. New York: Plenum, 1979.
- Woodring, S. F., C. D. Rossiter, and B. J. Yates. Pressor response elicited by nose-up vestibular stimulation in cats. *Exp. Brain Res.* 113: 165-168, 1997.
- Yates, B. J. Vestibular influences on the sympathetic nervous system. *Brain Res. Rev.* 17: 51-59, 1992.
- Yates, B. J. Vestibular influences on the autonomic nervous system. *Ann. NY Acad. Sci.* 781: 458-473, 1996.
- Yates, B. J., C. D. Balaban, A. D. Miller, K. Endo, and Y. Yamaguchi. Vestibular inputs to the lateral tegmental field of the cat: potential role in autonomic control. *Brain Res.* 689: 197-206, 1995.
- Yates, B. J., T. Goto, and P. S. Bolton. Responses of neurons in the rostral ventrolateral medulla of the cat to natural vestibular stimulation. *Brain Res.* 601: 255-264, 1993.
- Yates, B. J., L. Grélot, I. A. Kerman, C. D. Balaban, J. Jakus, and A. D. Miller. Organization of vestibular inputs to nucleus tractus solitarius and adjacent structures in cat brain stem. *Am. J. Physiol.* 267 (*Regulatory Integrative Comp. Physiol.* 36): R974-R983, 1994.
- Yates, B. J., J. Jakus, and A. D. Miller. Vestibular effects on respiratory outflow in the decerebrate cat. *Brain Res.* 629: 209-217, 1993.
- Yates, B. J., and A. D. Miller. Properties of sympathetic reflexes elicited by natural vestibular stimulation: implications for cardiovascular control. *J. Neurophysiol.* 71: 2087-2092, 1994.
- Yates, B. J., Y. Yamagata, and P. S. Bolton. The ventrolateral medulla of the cat mediates vestibulosympathetic reflexes. *Brain Res.* 552: 265-272, 1991.