

# Influences of neck afferents on sympathetic and respiratory nerve activity

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**ABSTRACT:** It is well established that the vestibular system influences the sympathetic nervous system and the respiratory system; presumably, vestibul sympathetic and vestibulorespiratory responses participate in maintaining stable blood pressure and blood oxygenation during movement and changes in posture. Many brainstem neurons that generate vestibulospinal reflexes integrate signals from the labyrinth and neck muscles to distinguish between head movements on a stable body and whole body movements. In the present study, responses were recorded from the splanchnic (sympathetic), hypoglossal (inspiratory) and abdominal (expiratory) nerves during stimulation of the C2 dorsal root ganglion or C2 or C3 nerve branches innervating dorsal neck muscles. Stimulation of neck afferents using low current intensities, in many cases less than twice the threshold for producing an afferent volley recordable from the cord dorsum, elicited changes in sympathetic and respiratory nerve activity. These data suggest that head rotation on a stable body would elicit both cervical and vestibular inputs to respiratory motoneurons and sympathetic preganglionic neurons. The effects of cervical afferent stimulation on abdominal, splanchnic and hypoglossal nerve activity were *not* abolished by transection of the brainstem caudal to the vestibular nuclei; thus, pathways in addition to those involving the vestibular nuclei are involved in relaying cervical inputs to sympathetic preganglionic neurons and respiratory motoneurons. Transection of the C1-3 dorsal roots enhanced responses of the splanchnic and abdominal nerves to pitch head rotations on a fixed body but diminished responses of the hypoglossal nerve. Thus, neck and vestibular afferent influences on activity of respiratory pump muscles and sympathetic outflow appear to be antagonistic, so that responses will occur during whole body movements but not head movements on a stationary trunk. In contrast, neck and vestibular influences on tongue musculature are complementary, presumably to produce tongue protrusion either during movements of the head alone or of the whole body. © 1999 Elsevier Science Inc.

**KEY WORDS:** Neck, Vestibular, Muscle spindle, Golgi tendon organ, Respiration, Sympathetic nervous system, Blood pressure.

## INTRODUCTION

There is considerable evidence to suggest that the vestibular system influences activity of both sympathetic and respiratory nerves

in the cat [reviewed in 25,26,29]. Electrical stimulation of the vestibular nerve results in changes in activity of a number of sympathetic nerves [24], as well as nerves innervating the diaphragm, intercostal muscles and abdominal muscles [27], and nerves innervating muscles regulating upper airway resistance [19]. Natural stimulation of vestibular receptors, produced by head rotations (*on a fixed body*) in animals with extensive denervations (including transections of cervical dorsal roots and the IXth and Xth cranial nerves) to remove nonlabyrinthine inputs that could be elicited by head movements, also has prominent effects on the cardiovascular and respiratory systems. Nose-up rotation of the head produces increases in sympathetic nerve activity [28] and an increase in blood pressure of approximately 20 mm Hg [23]. The same stimulus also produces large increases in activity of nerves innervating abdominal muscles [16] and tongue musculature [17] and less pronounced augmentation of discharges in the phrenic nerve, which innervates the diaphragm [16]. It has been hypothesized that vestibular influences on the sympathetic nervous system serve to offset orthostatic hypotension, which can result during nose-up body rotations in quadrupeds [25,26]. Vestibular-respiratory reflexes presumably contribute to adjustments in breathing and airway patency during changes in posture and also could be directly involved in producing movement and postural change [29].

Vestibular inputs to the brainstem are elicited whenever the head changes position in space, either when the head is turned on a stationary body or when a whole body movement occurs. Many reflexes elicited by vestibular stimulation, such as vestibulospinal reflexes acting on the limbs, are necessary only during whole body movements (i.e., when the body changes position in space, as during falling) but not when the head is turned on the body. Thus, the central nervous system must distinguish between whole body movements and head movements with the trunk fixed in space in generating appropriate compensatory responses [22]. For this reason, many vestibular nucleus neurons, including those that mediate vestibulospinal reflexes, receive convergent signals from vestibular and neck receptors with respective orientation vectors pointing in opposite directions. A head tilt that produces an excitatory vestibular response in these neurons would also produce an inhibitory neck response [2,8]. The gain components of neck and

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vestibular influences on limb muscles are also similar, so that during head movements on a stationary body, the opposing neck and vestibular inputs will cancel each other [2,8,21]. However, neck and vestibular influences are not antagonistic for all vestibular reflexes. Vestibular reflexes acting on neck muscles (vestibulo-collic reflexes) and reflex contractions of neck muscles elicited by stimulation of neck proprioceptors (cervicocollic reflexes) complement each other [3,13]. The addition of vestibulo-collic and cervicocollic reflexes results in stability of head position.

The prospect that inputs from the neck might influence brainstem systems that control respiration and circulation is yet to be considered. The present study had three components. In one group of experiments, electrical stimulation of either the C2 dorsal root ganglion or C2 and C3 nerve branches innervating the dorsal neck muscles biverter cervicis and complexus was used to activate neck afferents; the current intensities and number of shocks delivered were varied to determine the class (Group I–IV) of afferents influencing activity of the hypoglossal, abdominal and greater splanchnic (sympathetic) nerves. In a subset of these experiments, the responses to cervical afferent stimulation were recorded before and after transection of the brainstem 1–3 mm rostral to the obex, to separate portions of the vestibular nuclei that mediate vestibulo-sympathetic and vestibulorespiratory responses (located 4–5 mm rostral to the obex) [16,19,27,28] from respiratory motoneurons and sympathetic preganglionic neurons. The aim of this experiment was to determine whether the vestibular nuclei are a necessary part of the neural pathway that mediates cervicosympathetic and cervicorespiratory responses. In an additional group of animals, responses to sinusoidal or static pitch rotations of the head on a fixed body were recorded before and after transection of the upper cervical dorsal roots, so that we could determine whether neck and vestibular influences on sympathetic and respiratory outflow are antagonistic or complementary.

## MATERIALS AND METHODS

The procedures used in this study conformed with the National Institutes of Health Guidelines for Care and Use of Animals and were approved by the University of Pittsburgh Animal Care and Use Committee.

### *General Surgical Procedures*

Experiments were performed on 24 adult cats of either sex. Anesthesia was induced and maintained with 2–4% halothane (Fluothane, Ayerst Laboratories) vaporized in NO<sub>2</sub> and O<sub>2</sub>. Blood pressure was monitored from a femoral artery (with the use of Millar Mikro-tip transducer), and rectal temperature was monitored and maintained between 36 and 38°C with the use of an infrared lamp and heating pad. A tracheostomy was performed, and both femoral veins were cannulated to permit intravenous injections. If necessary, an intravenous infusion of Aramine (metaraminol bitartrate, Merck, Sharpe and Dohme, 80 µg/mL) was used to keep mean blood pressure >100 mm Hg. The animal's head was secured in a stereotaxic frame, and the body was also supported by hip pins and a clamp attached to the dorsal process of the T1 vertebra. The common carotid arteries were ligated, a midcollicular decerebration was performed, anesthesia was removed and paralysis was produced with gallamine triethiodide (Sigma; initial injection of 10 mg/kg i.v., which was supplemented by hourly injections of 5 mg/kg i.v.). While paralyzed, animals were artificially respired using a positive pressure ventilator; end-tidal CO<sub>2</sub> was monitored and maintained near 4% (range, 3.5–4.5%).

At the conclusion of the recording session, animals were eu-

thanzed using an overdose of sodium pentobarbital (120 mg/kg injected i.v.).

### *General Recording Procedures*

In each animal, one or more of the following nerves were dissected for recording: the hypoglossal nerve, an L1 or L2 spinal nerve trunk innervating abdominal muscles (internal oblique, transverse abdominus, external oblique and rectus abdominus) and the greater splanchnic nerve. The hypoglossal nerve was inserted into a bipolar silver tunnel electrode, and the splanchnic and abdominal nerves were placed on bipolar silver hook electrodes. Nerves prepared for stimulation or recording were covered with either warm mineral oil or a mixture of warm mineral oil and petroleum jelly.

Activity recorded from the hypoglossal, splanchnic or abdominal nerves was amplified by a factor of 10,000–500,000, filtered with a bandpass of 10–10,000 Hz, full wave rectified and integrated with a time constant of 1 ms. The signals were sampled at 500 Hz, averaged, stored and displayed with the use of a Cambridge Electronic Design (CED) 1401-plus data collection system interfaced with a Macintosh Quadra 800 computer.

### *Electrical Stimulation of Cervical Afferents*

In 15 animals, the C2 dorsal root ganglion was isolated on one side by reflecting and removing the overlying muscles [see 14 for anatomic details]. In 13 animals, branches of the C2 or C3 dorsal rami that clearly terminated in the dorsal neck muscles biverter cervicis and complexus were dissected free and prepared for electrical stimulation. The C2 dorsal root ganglion and/or muscle branches of the dorsal rami were mounted on bipolar silver electrodes for stimulation. Responses were recorded from the abdominal, splanchnic and hypoglossal nerves during electrical stimulation of cervical afferents, as described above. Sufficient sweeps were collected to obtain a clear response to nerve stimulation; typically, about 200 waveforms were averaged.

A small hemilaminectomy was performed to expose the dorsolateral portion of the C2 and C3 spinal cord ipsilateral to the nerve branches or dorsal root ganglion exposed for stimulation. The dura matter was opened, and a silver ball electrode was placed on the dorsal root entry zone of the C2 or C3 spinal cord segment for the purpose of recording compound action potentials elicited by cervical afferent stimulation. Afferent volleys were amplified by a factor of 1,000–10,000 and filtered with a bandpass of 10–10,000 Hz but were not rectified or integrated. Nerve volleys were collected (at 10,000 Hz), averaged and displayed using the CED data collection system and Macintosh computer described above.

The C2 dorsal root ganglion or muscle branches of the C2 or C3 dorsal ramus were stimulated using square wave current pulses. Both single shocks and trains of up to five shocks (interpulse interval of 3 ms) were used; the shock duration was 0.15 ms. When stimulating cervical afferents, we initially used current intensities that were five times the threshold (T) necessary to produce an afferent volley recordable from the cord dorsum. Subsequently, both higher and lower strengths were used. We also determined the minimal number of shocks required to elicit a response using a stimulus strength that was five times that necessary to produce an afferent volley.

### *Removal of Cervical Inputs to the Vestibular Nuclei*

In six experiments, responses were recorded from respiratory and sympathetic nerves during stimulation of cervical afferents before and after transecting the brainstem 1–3 mm rostral to the

obex. The brainstem was transected to test the hypothesis that connections between the vestibular nuclei and sympathetic preganglionic neurons and respiratory motoneurons are not necessary for the production of cervicosympathetic and cervicorepiratory reflexes. At the conclusion of these experiments, the brainstem was removed, fixed in formalin, cut into 100- $\mu\text{m}$  transverse sections and stained with Thionin so that we could verify that the brainstem transections were complete and could determine their locations.

#### Recording of Responses to Natural Vestibular Stimulation

In four animals, responses were recorded from sympathetic and respiratory nerves during sinusoidal or static head rotations in the pitch plane before and after transection of the C1-3 dorsal roots. Head rotations were produced as described in previous publications [16,17,28]. The animal's head was pitched-down 30° and supported by a skull-mounted cylinder that was attached to a head rotator that could move the head in the pitch plane. The pitch axis passed approximately through the center of the skull-C1 joint. The glossopharyngeal and vagus nerves were cut in experiments using head rotation, so that we were assured that visceral inputs elicited by head movement did not produce changes in respiratory or sympathetic nerve activity. Sinusoidal pitch rotations (20° amplitude) were delivered at 0.1–0.5 Hz, and static nose-up head rotations were delivered at 50° amplitudes. After responses to initial head rotations were recorded, a C1-4 laminectomy was performed, and the C1-3 dorsal roots were transected bilaterally. The amplitude of responses to head rotation before and after the upper cervical dorsal rhizotomy were compared to determine whether neck and vestibular influences on respiratory and sympathetic outflow are complementary or antagonistic.

Responses to sinusoidal natural vestibular stimulation were fitted with a sine wave using a least-squares method, and the response gain was calculated as described in previous publications [16,28]. The amplitude of responses to static head rotation was determined by dividing the mean nerve activity during the time the head was held nose-up by the mean nerve activity over a similar duration of time immediately after the tilt.

## RESULTS

In 15 experiments, respiratory or sympathetic nerve responses to stimulation of the C2 dorsal root ganglion were recorded. The mean minimal current intensity required to produce an afferent volley recordable from the cord dorsum was  $19 \pm 2$  (SEM)  $\mu\text{A}$ ; the median current intensity was 20  $\mu\text{A}$  (range, 10–30  $\mu\text{A}$ ). In 13 experiments, responses to stimulation of branches of the C2 or C3 dorsal rami innervating dorsal neck muscles were recorded. The mean threshold for producing an afferent volley was  $15 \pm 1$   $\mu\text{A}$ ; the median threshold was 15  $\mu\text{A}$  (range, 10–20  $\mu\text{A}$ ).

#### Splanchnic Nerve Responses to Afferent Stimulation

In six animals, responses of the splanchnic nerve to stimulation of the C2 dorsal root ganglion were recorded; in eight animals, responses to stimulation of branches of the C2 or C3 dorsal rami innervating dorsal neck muscles were examined. Table 1 indicates the thresholds of these responses, relative to the minimum current intensity required to produce an afferent volley recordable from the cord dorsum. In all cases, stimulation of cervical afferents at intensities  $<5T$  produced a change in splanchnic nerve activity; in about half the animals, the threshold was demonstrated to be  $<2T$ . Similar responses were elicited by stimulation of the ipsilateral or contralateral side.

TABLE 1  
THRESHOLDS FOR PRODUCING RESPONSES IN THE SPLANCHNIC, HYPOGLOSSAL AND ABDOMINAL NERVES, RELATIVE TO THE MINIMUM CURRENT INTENSITIES REQUIRED TO ELICIT AN AFFERENT VOLLEY RECORDABLE FROM THE CORD DORSUM

Nerve	1–2T	2–5T	>5T	<5T*
<i>Stimulation of C2 Dorsal Root Ganglion</i>				
Splanchnic nerve	3/6	2/6		1/6
Hypoglossal nerve	3/10	2/10	1/10	4/10
Abdominal nerve	2/6	3/6	1/6	
<i>Stimulation of Branches of C2 or C3 Dorsal Ramus Innervating Dorsal Neck Muscles</i>				
Splanchnic nerve	3/8	1/8		4/8
Hypoglossal nerve	7/10			3/10
Abdominal nerve	1/5	2/5		2/5

Ratios indicate the number of animals in which each range of thresholds was noted.

\* Responses were determined to have a threshold  $<5T$ , but the threshold range was not determined more precisely.

Figure 1A shows examples of sympathetic nerve responses to C2 dorsal root ganglion stimulation, whereas Fig. 1B shows responses of the splanchnic nerve to stimulation of C2 nerve branches innervating dorsal neck muscles. The splanchnic nerve

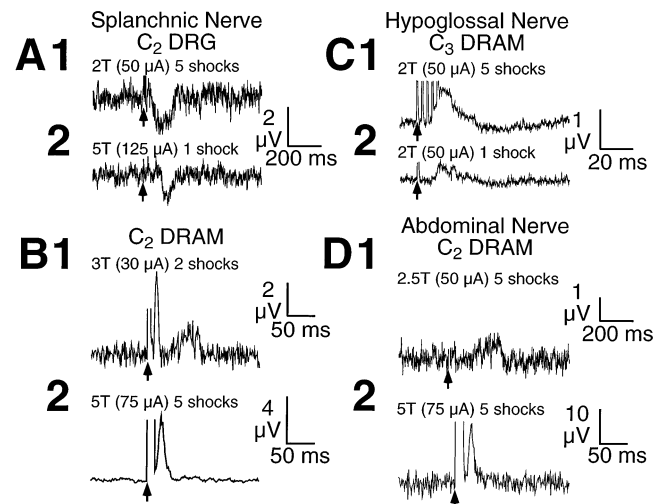


FIG. 1. Responses of respiratory and sympathetic nerves to electrical stimulation of neck afferents. (A) Examples of responses of the splanchnic nerve to low-threshold stimulation of the C2 dorsal root ganglion (DRG). Both records A1 and A2 were obtained from the same animal. (B) Splanchnic nerve responses to stimulation of a branch of the C2 dorsal ramus (DRAM) innervating neck muscles; the two traces (B1 and B2) in this panel are from *different* animals. (C) Two examples (waveforms C1 and C2) from the same animal of hypoglossal nerve responses to stimulation of the C3 DRAM. (D) Abdominal nerve responses to stimulation of muscle afferents. Traces D1 and D2 are from *different* animals. For all traces, the stimulus intensity used, both absolute current intensity and the threshold (T) relative to the current intensity required to produce an afferent volley recordable from the cord dorsum, is indicated. Arrows at the bottom of each panel indicate the latency of the first shock in the stimulus train. The responses are averages of 300–500 sweeps.

TABLE 2  
LATENCY AND DURATION OF RESPONSES ELICITED BY  
STIMULATION OF CERVICAL AFFERENTS

Nerve	Latency (ms)*		Duration (ms)†	
	Mean	SEM	Mean	SEM
<i>Stimulation of C2 Dorsal Root Ganglion</i>				
Splanchnic nerve	64.4‡	22.2	125.0	53.2
Hypoglossal nerve	16.7	0.5	21.8	7.9
Abdominal nerve	18.8	1.4	77.0	41.7
<i>Stimulation of Branches of C2 or C3 Dorsal Ramus Innervating Dorsal Neck Muscles</i>				
Splanchnic nerve	98.8	36.5	103.3	51.9
Hypoglossal nerve	12.7	2.1	31.0	17.1
Abdominal nerve	21.1	3.9	141.7	27.7

\* Measured from the first stimulus of a five-shock train.

† Determined from responses elicited by a five-shock stimulus at 5T.

‡ Significantly different from other values ( $p < 0.05$ ) by one-way ANOVA combined with the Tukey-Kramer Multiple Comparisons Test.

responses to cervical afferent stimulation consisted of a long-latency component (e.g., panel A2 of Fig. 1), a short-latency component (e.g., panel B2 of Fig. 1) or more typically of both short- and long-latency components (e.g., panel B1 of Fig. 1). Stimulation of hindlimb afferents has also been shown to produce short- and/or long-latency components in sympathetic nerves [18]. Table 2 indicates the latency and duration of splanchnic nerve responses to cervical afferent stimulation. These values had a large SEM, due to the fact that a short-latency component was not present in every animal.

#### Hypoglossal and Abdominal Nerve Responses to Neck Afferent Stimulation

In 10 animals, hypoglossal nerve responses to stimulation of the C2 dorsal root ganglion and nerve branches innervating dorsal neck muscles were recorded. Examples of hypoglossal nerve responses elicited by stimulation of muscle nerve branches are shown in Fig. 1C. Similar responses were elicited by stimulation of the ipsilateral and contralateral side, and (as indicated in Table 1) effects could be produced by stimulus intensities  $<5T$  (and typically  $<2T$ , particularly when stimulating nerve branches innervating dorsal neck muscles). An ANOVA showed that the latency of the hypoglossal nerve response to C2 dorsal root ganglion stimulation was significantly shorter than the latency of the response of the splanchnic nerve.

In six experiments, abdominal nerve responses to stimulation of the ipsilateral or contralateral C2 dorsal root ganglion were recorded, and in five animals we studied responses to stimulation of C2 and C3 nerve branches innervating dorsal neck muscles. Examples of abdominal nerve responses to stimulation of cervical muscle afferents are illustrated in Fig. 1D. The abdominal nerve responses to cervical afferent stimulation were similar to those recorded from the hypoglossal nerve in that comparable effects could be elicited by stimulation on either side, the changes in nerve activity typically had thresholds  $<5T$  (Table 1) and responses elicited by C2 dorsal root ganglion stimulation had significantly shorter latencies than those recorded from the splanchnic nerve (Table 2).

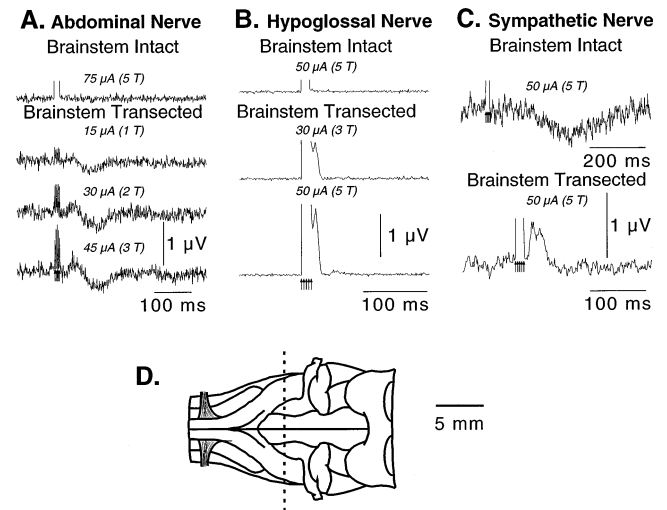


FIG. 2. Effect of a transection through the caudal medulla on abdominal (A), hypoglossal (B) and sympathetic (C) nerve responses to stimulation of low-threshold afferents from cervical muscles. D shows the approximate location of the brainstem transection in these experiments. Note that in C, the bottom trace is depicted on a different time scale than the top trace so that the short-latency response component is more obvious. As in Fig. 1, responses are the average of approximately 300–500 sweeps, and arrows at the bottom of each panel indicate the latency of stimuli. All examples were elicited by a train of five stimuli.

#### Effects of Brainstem Transection on Responses to Electrical Stimulation of Cervical Afferents

In six animals, we compared responses of the splanchnic, hypoglossal or abdominal nerves with stimulation of C2 or C3 nerve branches innervating dorsal neck muscles before and after transection of the brainstem. The brainstem transections were histologically confirmed to be complete and to be located 1–3 mm rostral to the obex. In all six animals, splanchnic nerve responses to neck afferent stimulation were recorded. In two experiments, a short-latency component (latency  $<30$  ms from first shock in the stimulus train) was prevalent in the response, but in the other four animals the response latency was  $>60$  ms. In both animals with a short-latency response, this component was unaffected by the brainstem transection. However, in all cases, long-latency components in the splanchnic nerve responses were abolished by sectioning the brainstem. In three animals, a short-latency response replaced the long-latency component after brainstem transection, as illustrated in Fig. 2C. These findings suggest that whereas neck afferent inputs can reach sympathetic preganglionic neurons through pathways in addition to those involving the vestibular nuclei, neurons located in the rostral medulla or pons are critical for producing long-latency components of cervico-sympathetic responses.

In five animals, effects of a midmedullary brainstem transection on hypoglossal nerve responses to cervical afferent stimulation were examined. In all five cases, the responses persisted after sectioning of the brainstem. In one experiment, the response amplitude increased after the transection, as illustrated in Fig. 2B. These results show that the vestibular nuclei are not an essential part of the neural pathway relaying cervical inputs to hypoglossal motoneurons.

In four experiments, the effects of brainstem transection on cervicoabdominal responses were determined. In all four animals,

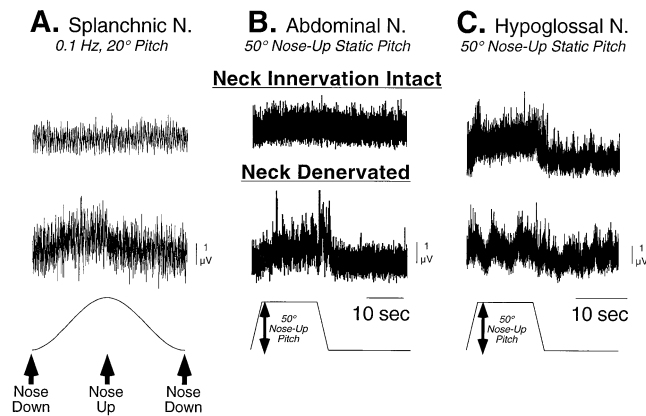


FIG. 3. Responses of sympathetic and respiratory nerves to head rotation, before and after transection of the C1-3 dorsal roots. (A) Responses of the splanchnic nerve to 20° sinusoidal pitch head rotation; traces are the average of approximately 250 sweeps. (B) Responses of the abdominal nerve to 50° static nose-up rotation of the head. (C) Hypoglossal nerve responses to static nose-up head rotation. After the dorsal rhizotomy, spontaneous respiratory-related discharges in the hypoglossal nerve increased, resulting in noisier background activity than when the cervical dorsal roots were intact. The traces in B and C are the average of approximately 25 sweeps.

the lesion resulted in a pronounced *increase* in response amplitude, as illustrated in Fig. 2A. This observation suggests that the transmission of cervical inputs to abdominal motoneurons in the decerebrate cat is tonically inhibited by neurons located in the rostral medulla or pons.

#### *Integrated Influences of Cervical and Vestibular Inputs to Respiratory Motoneurons and Sympathetic Preganglionic Neurons*

In four animals, responses of the hypoglossal, abdominal and splanchnic nerves to head rotations in the pitch plane were recorded before and after transection of the C1-3 dorsal roots. Little response was elicited in the splanchnic nerve during sinusoidal or static head rotations in animals with an intact neck innervation; however, transection of the upper cervical dorsal roots unmasked the response (e.g., see Fig. 3A). The average response amplitudes after cutting the upper cervical dorsal roots were  $453 \pm 247\%$  of those when neck inputs were present. In other words, sympathetic nerve responses to pitch head rotations when neck inputs were present (i.e., when the cervical dorsal roots were intact) were on average  $<25\%$  of the amplitude of responses when neck inputs were removed (i.e., after the cervical dorsal rhizotomy). Thus, neck and vestibular influences on the sympathetic nervous system appear to be integrated in an antagonistic fashion, so that powerful changes in sympathetic outflow will occur during whole body movements but not during head rotations on a stationary body that elicit both vestibular and neck signals.

In a similar fashion, inputs from cervical receptors antagonized vestibular influences on abdominal nerve activity during head rotation; the mean response amplitude after the cervical dorsal rhizotomy was  $463 \pm 349\%$  of that when neck inputs were present (see Fig. 3B for illustration). In contrast, powerful hypoglossal nerve responses to head rotation were present in animals with an intact neck innervation. Transection of the upper cervical dorsal roots *reduced* the amplitude of the hypoglossal nerve responses to

static head pitch to  $36.4 \pm 19.8\%$  of that when neck inputs were present. These observations suggest that neck and vestibular influences on hypoglossal motoneurons during pitch rotations are complementary.

## DISCUSSION

The present study shows that cervical afferents, including those from dorsal neck muscles, influence activity of sympathetic preganglionic neurons, abdominal motoneurons and hypoglossal motoneurons. The cervical afferents producing changes in sympathetic and respiratory outflow include muscle afferents that can be activated by stimulus intensities less than twice the threshold for eliciting an afferent volley recordable from the cord dorsum. In the hindlimb of the cat, such low stimulus strengths have been demonstrated only to excite afferents from muscle spindles and Golgi tendon organs [4,5]. This is also presumably the case for afferents from dorsal neck muscles, because some neck spindle afferents have high conduction velocities of up to 90 m/s and neck Golgi tendon organ afferents have conduction velocities between 50 and 70 m/s [1]. However, muscle spindle afferents from the neck are also unlike those from the hindlimb, in that some neck spindle afferents have slow conduction velocities of between 10 and 40 m/s [15]. Thus, low-threshold stimulation of nerves innervating dorsal neck muscles may only activate a small number of afferents. This could explain why stimulation of neck muscle nerves at no more than two times the threshold for producing an afferent volley did not produce a perceptible change in sympathetic and respiratory nerve activity in all animals. It is conceivable that in the cases where low-intensity stimulation was ineffective, an insufficient number of afferents was activated to produce a large enough change in sympathetic or respiratory outflow to be detected in recordings from peripheral nerves. The observation that responses in the abdominal nerve were enhanced by a transection through the caudal medulla also suggests that in some cases the lowest threshold responses in this nerve could have been masked by tonic inhibition from the rostral medulla or pons. It is possible that removal of the inhibitory descending signals may have resulted in the low-threshold responses being more obvious in some experiments.

A number of previous studies have considered the effects of inputs from hindlimb afferents on sympathetic nerve activity in the cat. These studies showed that low-threshold stimulation of hindlimb cutaneous or mixed nerves elicits changes in sympathetic outflow [7,18]; this finding was confirmed by the present study. In contrast, stimuli applied to hindlimb muscles or muscle nerves must activate small-diameter afferents (and not hindlimb spindle or Golgi tendon organ afferents) to produce changes in sympathetic and respiratory nerve activity [9–11,20]. However, the present study showed that stimulation of muscle branches of cervical dorsal rami at intensities  $<5T$  (and usually at much lower intensities) produced changes in sympathetic and respiratory nerve activity; this stimulus strength should have mainly excited spindle and Golgi tendon organ afferents [1,4,5]. Thus, inputs from large neck and large hindlimb muscle afferents may have different effects on the sympathetic nervous system and the respiratory system. Although this finding remains to be definitively confirmed by studies that selectively stimulate neck muscle spindles and Golgi tendon organs using methods such as vibration and injection of succinylcholine, it is not surprising. It is well established that stimulation of the vestibular system can affect activity in sympathetic and respiratory nerves [24–26,29]; these responses are mediated by neurons in the medial and inferior vestibular nuclei [27]. Many neurons in the vestibular nuclei receive convergent inputs from the labyrinth and neck muscle spindle afferents [2,8]; inputs

from the neck to vestibulospinal neurons allow these cells to discriminate between head movements on a stationary body and whole body movements. Presumably, inputs from the neck to sympathetic preganglionic neurons and respiratory neurons are relayed in part through cells in the vestibular nuclei with convergent neck and vestibular signals.

However, these experiments showed that neck inputs are also relayed to sympathetic and respiratory neurons through circuits that do not involve the vestibular nuclei, as demonstrated by the fact that stimulation of nerve branches innervating dorsal neck muscles produced effects in the splanchnic, hypoglossal and abdominal nerves in animals with a transection through the medulla. On the other hand, although cervicorespiratory and cervicosympathetic responses were *present* in animals without functional connections from the rostral medulla and pons to respiratory motoneurons and sympathetic preganglionic neurons, these effects were altered by a midmedullary transection. For example, long-latency components of splanchnic nerve responses to head rotation were abolished by the lesion. Previous studies have shown that neurons in the rostral ventrolateral medulla relay vestibular and other somatic inputs, including those from the hindlimb, to sympathetic preganglionic neurons [12,30]. It is thus possible that elimination of descending projections from the rostral ventrolateral medulla to the spinal cord, and not the vestibular nuclei, was responsible for the effects of the medullary transection on cervicosympathetic responses. In addition, brainstem transection enhanced cervicoabdominal responses, indicating that tonic inhibition from the rostral medulla or pons suppresses these effects. The location of the neurons providing this inhibition is yet to be determined but could potentially include cells in the vestibular nuclei.

These data thus suggest that the neural pathways mediating cervicosympathetic and cervicorespiratory responses may be very complex, potentially involving connections through the vestibular nuclei and circuits only including the caudal brainstem and spinal cord. The physiological significance of these multiple pathways conveying cervical influences on respiratory and sympathetic outflow remains to be discerned.

Another group of experiments compared sympathetic and respiratory nerve responses to head rotation before and after elimination of cervical inputs through transection of the C1-3 dorsal roots. These experiments indicated that neck and vestibular influences on respiratory pump muscles and the sympathetic nervous system cancel each other, as in the case of vestibulospinal reflexes [21]. This response pattern is functionally appropriate, so that a compensatory response to maintain stable blood pressure or blood oxygenation will only occur when the body changes position in space and not when the head moves but the body remains stationary. In contrast, neck and vestibular inputs to hypoglossal motoneurons appear to add with each other (i.e., they are complementary), as in the case of vestibulocollic reflexes [3,13]. This response pattern is physiologically adaptive, because an increase in activity of tongue protruder muscles is required to maintain airway patency during either a nose-up rotation of the whole body or a nose-up rotation of the head on a stationary body. The differences in the integration of neck and vestibular influences on respiratory pump muscles and upper airway muscles underscores the complexity of vestibuloautonomic responses and indicates that relatively complex pathways must mediate these responses.

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