Medial vestibular nucleus mediates the cardiorespiratory responses to fastigial nuclear activation and hypercapnia

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Medial vestibular nucleus mediates the cardiorespiratory responses to fastigial nuclear activation and hypercapnia. J Appl Physiol 97: 835–842, 2004; 10.1152/japplphysiol.00134.2004.—Electrical stimulation of the cerebellar fastigial nucleus (FN) evokes hyperventilation and hypertension responses that are similar to those induced by stimulation of the medial region of the vestibular nucleus (VN M). Because there are mutual projections between these two nuclei morphologically, we hypothesized that the FN-mediated cardiorespiratory responses were related to the integrity of the VN M. Experiments were conducted on 21 anesthetized, tracheotomized, and spontaneously breathing rats. Electrical stimulation (~10 s) of the FN was used to evoke cardiorespiratory responses, and the same stimulus was repeated 30–45 min after bilateral lesions of the VN M by local micro-injection of ibotenic acid (100 mM, 100 nl). We found that FN stimulation-induced hyperventilation and hypertension were attenuated significantly by the lesions. The role of the VN M in the ventilatory responses to chemical challenges was subsequently defined. The animals were exposed to hypercapnia (10% CO₂) and hypoxia (10% O₂) for 1–2 min randomly before and after VN M lesions. The results showed that VN M lesions significantly attenuated the cardiorespiratory responses to hypercapnia but not to hypoxia, with little effect on baseline respiratory variables. These findings suggest that the VN M is required for full expression of the cardiorespiratory responses to electrical stimulation of the FN as well as to hypercapnia. However, neurons within the VN M do not appear to be critical for maintaining eupneic breathing and the cardiorespiratory responses to hypoxia.

IT IS GENERALLY ACCEPTED that the cerebellar nuclei regulate movement and posture by adjusting the output of the major descending motor system of the brain. In addition, there is accumulating evidence to show the involvement of the cerebellar fastigial nucleus (FN) in respiratory central control. Bassal and Bianchi (2) first reported an altered respiratory response to electrical stimulation of the FN in cats. Subsequent studies confirmed that electrical (2, 14, 31, 35, 36) or chemical activation (41) of FN neurons predominantly elevated ventilation usually associated with a pressor response in both anesthetized cats and rats. Ablation of this region attenuated the respiratory response to severe hypercapnia and hypoxia in the rat and cat (34, 38). Several investigators (8, 15, 37), using extracellular recordings, have demonstrated in alert and anesthetized animals that respiratory-modulated neuronal activity within the vestibular nucleus is altered or substantially diminished by lesion of the NGC (18, 12, 16) altered respiration in cats and rabbits. Subsequent studies showed that electrical stimulation of the VN M in the rat produced hyperventilation and hypertension that were eliminated or substantially diminished by lesion of the NGC (18, 42). Interestingly, these cardiorespiratory responses and their dependence on the NGC are similar to those elicited by FN stimulation (40). To date, the role of the VN M in FN-mediated cardiorespiratory responses remains unclear. In addition, although the involvement of the VN M in respiratory modulation has been suggested, its contributions to the respiratory responses to chemical challenges, such as hypercapnia and hypoxia, have not been defined.

The aim of this study was to determine the importance of the VN M in cardiorespiratory responses to FN activation. We hypothesized that destruction of VN M neurons would attenuate central chemoreceptors. Further investigation showed that microinjection of acetazolamide into the FN to produce focal tissue acidosis profoundly enhanced the ventilation in anesthetized rats, suggesting the presence of local chemosensitive neurons (39). With respect to the FN effenter projections, previous studies have pointed out that FN-mediated respiratory responses are independent of the pontine respiratory groups and Bötzing complex (45) but depend on the integrity of the gigantocellular nucleus (NGC) (42). In addition, FN stimulation alters respiratory-modulated neuronal activity within the nucleus tractus solitarius and nucleus ambiguous (35). Collectively, these results suggest that the FN exerts an excitatory role in cardiorespiratory modulation, especially in the ventilatory response to severe hypercapnia via its connections with the medullary respiratory network.

Mutual neuronal connections between the FN and vestibular nucleus (VN), particularly its medial region (VN M), have been described both morphologically and functionally in different species, including the rat, cat, and monkey (3–5, 11, 19, 21). For example, anatomic studies in animals have confirmed that the FN efferent projections to the VN M in cardiorespiratory responses remains unclear. In addition, although the involvement of the VN M in respiratory modulation has been suggested, its contributions to the respiratory responses to chemical challenges, such as hypercapnia and hypoxia, have not been defined.

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FN-mediated cardiorespiratory responses. To further clarify contributions of the VN M to respiratory chemoreflexes, we also examined the effects of lesion of the VN M in the respiratory responses to hypercapnia and hypoxia. We found that, in anesthetized and spontaneously breathing rats, electrical stimulation of the FN significantly increased minute ventilation (Vt) and arterial blood pressure, and these responses were markedly attenuated after bilateral lesions of VN M neurons via local microinjection of ibotenic acid (IA). We also found that lesions of VN M neurons attenuated the hyperventilation and hypotension induced by hypercapnia (10% CO₂-21% O₂-69% N₂) but had no effect on the cardiorespiratory responses to hypoxia (10% O₂-90% N₂). In addition, lesions of the VN M did not significantly alter baseline respiratory variables. These results suggest that the VN M is required for full expression of the cardiorespiratory responses to stimulation of the FN and systemic hypercapnia. However, neurons within the VN M appear to be not critical for maintaining eupneic breathing and cardiorespiratory responses to hypoxia.

METHODS

General animal procedure. The experimental protocols described in this study were approved by the Institutional Animal Care and Use Committee in compliance with the Animal Welfare Act and were in accordance with the National Institutes of Health policy on humane care and use of laboratory animals. The experiments were conducted in 21 tracheotomized and spontaneously breathing Sprague-Dawley male rats (300–400 g) initially anesthetized with Nembutal (50 mg/kg ip). The left femoral vein and artery were cannulated, the former for anesthetic administration and the latter for monitoring arterial blood pressure and heart rate (HR). Appropriate supplemental anesthesia (chloralose and urethane; 100 mg/kg and 500 mg/kg, respectively), as needed, was administered intravenously to suppress corneal and withdrawal reflexes. The trachea below the larynx was tracheotomized by blunt dissection and cannulated with a tracheal cannula connected to a one-way breathing valve. The tracheal pressure was recorded via a pressure transducer that was connected to a side-port of the tracheal cannula. The core temperature was monitored with a rectal probe and maintained at ~37.5°C by a heating pad and radiant heat. Respiratory flow was measured with a pneumotachograph and a differential pressure transducer (model ML141, ADInstruments, Castle Hill, Australia). The flow signal was integrated by PowerLab/8SP (ADInstruments, model ML785) to generate tidal volume (Vt). The pneumotachograph was made of stainless steel and had a linear flow-pressure relationship in the range of 0–20 ml/s and a flow resistance of 0.046 cmH₂O·ml⁻¹·s⁻¹, with a dead space of ~0.2 ml. A three-way switch was attached to the inspiratory inlet of the one-way breathing valve and used to manipulate the inhaled gas mixture. End-tidal pressures of O₂ and CO₂ were monitored via an infrared O₂-CO₂ analyzer (model 78356A, Hewlett Packard, Louisville, KY).

Occipital craniotomy. Animals were placed in a rigid metal frame with the head fixed in a stereotaxic apparatus (model 1404, David Kopf, Tujunga, CA). A hole (~8 mm diameter) was drilled at the midline (11.5 mm posterior to the bregma) for stereotaxically inserting the electrode into the FN and a microneedle into the VN M, according to the rat brain atlas (22). Bleeding was controlled with bone wax, absorbable hemostat (Surgicel, Ethicon, Johnson & Johnson, Somerville, NJ), and the use of a bipolar coagulator (model 440S, Radionics, Burlington, MA). The underlying tissue, covered by a 2 x 2 sponge gauze, was saturated with mineral oil to prevent drying.

Electrical stimulation of the FN. To evaluate baseline cardiorespiratory variables became stable for at least 15 min, the following studies were conducted in 20 rats. Stereotaxic coordinates were used to unilaterally position (insert angle = 90°) a stainless steel, concentric bipolar electrode (model NE-100, Rhodes Medical Instruments, Woodland Hills, CA) into the right FN. The stimulating electrode was placed and fixed in a site where reproducible respiratory responses were clearly detectable during electrical stimulation. The electrode placement was 2.6–2.8 mm rostral to the obex, 0.5–1.5 mm lateral to the midline, and 5.5–6.5 mm from the surface of the cerebellar cortex. The stimulating parameters were delivered from a stimulator (model S88, Grass, Quincy, MA) at the beginning of either the inspiratory or expiratory phase. The stimulating intensity was fixed (200-ms trains of 0.2-ms pulses at 100 μA) throughout the experiment, whereas the stimulating frequency was varied (50, 100, 120, 150, and 200 Hz delivered for ~10 s) to find a threshold that evoked a detectable change in respiration. The placement site of the stimulating electrode was mapped on a grid.

Hypercapnic and hypoxic exposure. In 11 rats, hypercapnia (10% CO₂ mixed with 21% O₂ and 69% N₂) or acute hypoxia (10% O₂-balance N₂) for 1–2 min was induced, respectively, via turning a three-way switch from room air to one or the other stimulating mixed gases. These chemical challenges were performed after the FN electrical stimulation in 10 of 11 rats and without electrical stimulation in 1 rat. Stabilization of the baseline cardiorespiratory variables for at least 5 min was allowed before each chemical challenge.

Bilateral microinjection of IA in the VN M. After baseline cardiorespiratory responses were collected and measured for 3–5 s, the right VN M was anesthetized and spontaneously breathing rats, electrical stimulation in 10 of 11 rats and without electrical stimulation in 1 rat. Stabilization of the baseline cardiorespiratory variables for at least 5 min was allowed before each chemical challenge.

RESULTS

Cardiorespiratory responses to electrical stimulation of the FN. Either an excitatory (n = 15) or inhibitory (n = 5) ventilatory response to FN stimulation, defined by Ve, was observed when suprathreshold stimulation was applied, and
these responses usually lasted for 20–30 s. Among the excitatory responses, increases in both VT and f were observed in 11 rats, whereas in 4 rats f increase was coupled with either a slight decrease or no change in VT. In comparison, decreases in both VT and f were observed in the inhibitory ventilatory response. The latencies for the evoked excitatory and inhibitory ventilatory responses were 0.45 ± 0.03 and 0.46 ± 0.02 s, respectively (P > 0.05 between 2 types of responses). In all rats, these respiratory responses were followed by an associated pressor response that lasted <20 s, as previously reported (36, 40). The stimulating thresholds varied from 100 to 200 Hz, i.e., 100 Hz (n = 13), 120 Hz (n = 2), 150 Hz (n = 4), and 200 Hz (n = 1). Figure 1 displays typical cardiorespiratory responses to stimulation of the FN. As shown, electrical stimulation generated hyperventilation and pressor responses. Group data showing cardiorespiratory responses to FN stimulation are depicted in Fig. 2. VE was significantly increased by 45.4 ± 5.2% in 15 rats (Fig. 2A; P < 0.05) but decreased by 29.3 ± 6.1% in 5 rats (Fig. 2B; P < 0.05) compared with the control standardized as zero. These excitatory ventilatory changes were due to an elevation in both VT and f, whereas a decrease in VT was the dominant characteristic in inhibitory responses. A pressor response was always observed in both types of ventilatory responses (8.6 ± 3.5 and 5.8 ± 0.5%, P < 0.05). The electrodes were located within the FN as illustrated in Fig. 2C.

Effect of VN M lesions on the cardiorespiratory responses to electrical stimulation of the FN. We compared the cardiorespiratory responses to FN stimulation before and after IA injection into the VN M in 12 rats. Figure 3 exhibits a typical example of experimental recordings. As shown, the FN stimulation-induced hyperventilation and pressor responses (Fig. 3A, left) were eliminated 45 min after IA injection into the VN M (Fig. 3A, right). The injection was made within the VN M (Fig. 3B). Similar results were found in group data (Fig. 4). Two important points should be noted. First, VN M lesions attenuated the ventilatory responses to FN stimulation primarily via depression of VT response. Second, VN M lesions markedly diminished the cardiovascular responses to FN stimulation. The cardiorespiratory responses to bilateral injection of IA into the VN M were time dependent. As illustrated in Fig. 5, IA injection into the VN M initially significantly increased

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**Fig. 1.** Experimental recordings of the ventilatory and arterial blood pressure (ABP) responses to electrical stimulation of the fastigial nucleus (FN). Traces from top to bottom show ABP, air flow, tidal volume (VT), and tracheal pressure (Ptr), respectively. Arrows point to “on” and “off” of 100-Hz stimulation. Stimulation was <10 s.

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**Fig. 2.** Group data showing the effects of electrical stimulation of the FN on cardiorespiratory responses. The excitatory (A) and inhibitory (B) respiratory responses [minute ventilation (VT), respiratory frequency (f), and VT] coupled with the associated cardiovascular changes [mean ABP (MABP) and heart rate (HR)] are shown. Values are means ± SE; n = 15 and 5 for excitatory and inhibitory ventilatory responses, respectively. *P < 0.05 between the data obtained before (standardized as 0) and during electrical stimulation. C: placements of the electrodes referenced to stereotaxic coordinates and derived from electrode tracks are represented by ○ and ●, respectively (7 other sites are buried beneath these circles). IN and LN, interposed and lateral nuclei, respectively.
ventilation and HR without significant changes in mean arterial blood pressure (5 min after injection). Interestingly, 30–45 min after IA injection, baseline ventilation returned to control levels, but HR remained significantly higher than control. In three other rats, vehicle rather than IA was microinjected into the VN
M, and in one of the three rats a second vehicle injection was applied 120 min later. FN stimulation induced excitatory (2 trials) and inhibitory (2 trials) ventilatory responses. The absolute changes in $V_\dot E$ before (69.5 ± 26.4%) and after vehicle injection (62.0 ± 10.9%) were not significantly different. The relatively large ventilatory response as compared with those described in Fig. 2 is due to the small sample size in this control study.

Effect of VN
M lesions on the cardiorespiratory responses to hypercapnia and hypoxia. Hypercapnia and hypoxia were applied and compared immediately before and 30–45 min after local injection of IA into the VN
M in 11 rats. Hypercapnia significantly increased $V_t$, $f$, and $V_\dot E$ (Fig. 6A) but decreased mean arterial blood pressure with little effect on HR (Fig. 6B). The hyperventilatory response was significantly depressed by VN
M lesions predominantly via a reduction of $f$ response, and the hypotension in response to hypercapnia was decreased.

Fig. 3. A: experimental recordings of the cardiorespiratory responses to the FN stimulation before and after the medial region of the vestibular nucleus (VN
M) lesions. The responses before (left) and after (right) VN
M lesion are compared, in which the traces from top to bottom are ABP, air flow, $V_t$, and $Ptr$. Arrows point to on and off of 100-Hz stimulation. B: schematically illustrated representative areas stained by Chicago Sky Blue [ibotenic acid (IA)] injections. The numbers listed are the distances rostral to the obex. M, L, and S, vestibular medial, lateral, and spinal nuclei, respectively; Amb, ambiguous nucleus; and LPGi, lateral paragigantocellular nucleus.

Fig. 4. Group data presenting the effects of VN
M lesions on the cardiorespiratory responses to FN stimulation. Respiratory ($V_\dot E$, $f$, and $V_t$; A) and cardiovascular (MABP and HR; B) responses before and after VN
M lesions are compared. Values are means ± SE; $n = 12$. *$P < 0.05$ between the data obtained before and during electrical stimulation; †$P < 0.05$ between the responses before and after VN
M lesions.
FN is involved in respiratory modulations under pathophysiological conditions. Our observation that FN stimulation mainly produces an immediate ventilatory augmentation is consistent with previously reported evidence that the FN plays an excitatory role in respiratory modulation. First, electrical or chemical activation of FN neurons predominantly enhanced ventilation via elevation of VT and/or f (2, 14, 31, 35, 36, 41). Conversely, ablation of these neurons attenuated respiratory responses to chemical challenges, especially severe hypercapnia in the rat and cat (34, 38). Second, the FN contains both respiratory-modulated and chemosensitive neurons that participate in respiratory responses to severe chemical challenges (8, 15, 28, 37, 39). Third, respiratory-modulated neurons recorded in the nucleus tractus solitarius and nucleus ambiguus were responsive to electrical stimulation of the FN with an ~30 ms latency, suggesting that the FN projects into the medullary respiratory network via a fairly direct pathway. Compared with the neuronal response, a relatively longer latency for the ventilatory response to FN stimulation (~450 ms) was observed in the present study. This difference should include the signal transmission from the medullary respiratory-modulated neurons to the respiratory muscles. Identifying the FN involvement in modulation of chemoreception has significance in explaining pathological neuroanatomic findings in developmental disorders. For example, the FN has been established as one of the major regions in the central nervous system responding to hypercapnia (7) by utilizing functional magnetic resonance imaging. FN neuronal responses to hypercapnia were uniquely and significantly attenuated in patients with congenital central hypoventilation syndromes (6) and sleep-disordered breathing (10). In agreement, midline cerebellar damage-induced obstructive apnea during sleep was also observed in animals (1). Abnormalities, including severe gliosis, increased astrocytes, and neuronal immaturity, have been consistently observed in the cerebellar nuclei and cortex of the majority of sudden infant death syndrome victims (20, 33). In fact, abnormalities within the cerebellum, particularly in the FN, have been suggested to contribute to sudden infant death syndrome (9).

VN_M neurons are involved in the FN-mediated cardiorespiratory responses. One of our major findings is that, after selective destruction of VN_M neurons, the previously observed
hyperventilation and pressor responses to electrical stimulation of the FN were significantly attenuated. These results suggest that VN_M neurons are required for full expression of FN-mediated cardiorespiratory responses, supporting earlier reports. Morphologically, using retrograde or anterograde tracing techniques, the mutual projections between the FN and VN_M have been demonstrated in different species, including the rat, cat, and monkey (5, 11, 19, 21). Most of the projections from the FN to the VN_M are glutamatergic in the rat (5). Functionally, electrophysiological studies have shown that stimulation of the FN elicits monosynaptic excitation in vestibular neurons through FN projections (13). FN neurons receiving vestibular information are responsive to movements in space (26, 27), and both nuclei are involved in control of motor learning (25). Recently, results from functional magnetic resonance imaging studies suggest that the abnormalities of the efferent and afferent pathways of both vestibular and cerebellar nuclear structures may contribute to mechanisms underlying sudden infant death and a number of other sleep-disordered breathing syndromes (10). Our observation that VN_M neurons are required for full expression of the FN-mediated cardiorespiratory responses. Third, FN stimulation alters respiratory output and respiratory-modulated neuronal activity in the nucleus tractus solitarius and nucleus ambiguus by paucisynaptic connections because the latency was 10–60 ms. We cannot rule out the possibility that VN_M involvement in FN-mediated cardiorespiratory responses is due to its effects on the medullary respiratory neurons. In fact, activation of the VN via electrical stimulation of vestibular neurons modulates respiratory-modulated neuronal activity in these central respiratory groups (32, 46). Further studies are needed to clarify how CN neurons responsible for cardiorespiratory modulation are linked with the VN_M.

**How is the VN_M involved in FN-mediated cardiorespiratory responses?** The data obtained from the experiments reported here cannot answer this question directly. However, based on the morphological connections between the FN and other respiratory-related medullary nuclei, there are several possibilities that may help to interpret VN_M involvement. First, the presence of mutual projections between the FN and VN_M presents the possibility that efferent pathways of the FN responsible for cardiorespiratory modulation may partially pass through the VN_M and/or the FN and receive excitatory afferent inputs from the VN_M. Second, both hyperventilation and pressor responses to activation of the FN or VN_M apparently depend on the integrity of NGC neurons (18, 40). Therefore, VN_M neurons may have excitatory synaptic connections with those NGC neurons required for full expression of the FN-mediated cardiorespiratory responses. Third, FN stimulation alters respiratory output and respiratory-modulated neuronal activity in the nucleus tractus solitarius and nucleus ambiguus by paucisynaptic connections because the latency was 10–60 ms. We cannot rule out the possibility that VN_M involvement in FN-mediated cardiorespiratory responses is due to its effects on the medullary respiratory neurons. In fact, activation of the VN via electrical stimulation of vestibular neurons modulates respiratory-modulated neuronal activity in these central respiratory groups (32, 46). Further studies are needed to clarify how CN neurons responsible for cardiorespiratory modulation are linked with the VN_M.

**VN_M is involved in the cardiorespiratory responses to hypercapnia but not important to the cardiorespiratory responses to hypoxia.** To date, contributions of the VN_M to respiratory chemoreflexes have been poorly understood. Another major finding in the present study was that VN_M lesions significantly attenuated hyperventilation and hypotension in response to hypercapnia but failed to alter cardiorespiratory responses to acute hypoxia, suggesting that this structure is uniquely involved in the cardiorespiratory modulation during hypercapnia. Our finding of VN_M contribution to cardiorespiratory modulation, especially during hypercapnia, suggests that the function of the VN_M extends beyond vestibular action. The FN contains both chemosensitive (28, 39) and respiratory-modulated neurons (8, 15, 37), and lesions of these neurons profoundly attenuated the ventilatory response to hypercapnia (34). Therefore, it is possible that the involvement of VN neurons in hypercapnic ventilatory modulation is related to the presence of VN_M neurons. In support of this, enhanced apoptosis in sudden infant death syndrome victims was observed not only in the cerebellum but also in the VN_M (10). Moreover, the FN also plays a role in the ventilatory responses to airway mechanical and chemical stimulation (34). Therefore, the VN_M may also contribute to these FN-modulated respiratory responses.
Although the VN is involved in respiratory modulation, its subnucleus, VN\textsubscript{M}, seems not to be critical for eucapnic breathing. Electrical or chemical stimulation of the VN (2, 12, 16, 29), or especially the VN\textsubscript{M} (42), in the anesthetized rat, rabbit, and awake goat significantly altered ventilation. The VN contains respiratory-modulated neurons as demonstrated in studies on songbirds and budgerigars (23). Activation of the vestibular system in the cat via electrical stimulation of vestibular neurons or whole body tilt modulates firing behavior of respiratory-modulated neurons recorded in the pre-Bötzinger complex (46) and the medullary ventral respiratory group (32). These neurons have been suggested to be essential for maintaining respiratory rhythm and pattern. The functional significance of the VN in respiratory modulation has been addressed. Activation of the vestibular system by rotating the head in carotid-sinus-denervated, vagotomized, and decerebrate cats significantly altered respiration (24). More recently, studies carried out on humans indicated that stimulation of semicircular canals, but not otolith organs or neck muscle afferents, mediated increased ventilatory responses predominantly via elevation of f(17). In the present study, microinjection of IA into the VN\textsubscript{M} produced an immediate excitatory cardiorespiratory response. We believe that this response is due to the initial stimulating effect of IA on VN\textsubscript{M} neurons, consistent with previous reports in which microinjection of excitatory amino acids into the vestibular nuclei significantly altered cardiovascular activity (29, 42). The observation that ventilation did not significantly differ from baseline respiratory variables 30–45 min after IA injection supports the hypothesis that the VN\textsubscript{M} is not involved in respiratory modulation during eucapnic breathing. Interestingly enough, after VN\textsubscript{M} lesions, HR was still significantly higher than control, which suggests VN\textsubscript{M} involvement in cardiovascular modulation. In agreement, a role for the VN in cardiovascular modulation has been reported. For example, c-fos-like immunoreactivity was observed heavily in the VN during stimulation of the baroreceptors (30). Moreover, electrical or chemical stimulation of the VN, including the VN\textsubscript{M}, altered cardiovascular activity (29, 42).

Limitation of experimental methods. In general, cerebellar and vestibular structures exert substantial influences on breathing and cardiovascular activity, particularly under conditions of extreme challenges. The degree of hypoxia (10% O\textsubscript{2} for 1–2 min) that we used may be insufficient to demonstrate VN\textsubscript{M} influence on cardiorespiratory modulation. On the other hand, because the lesions were localized at the vicinity of the VN\textsubscript{M}, our results cannot account for the full function of the VN in cardiorespiratory modulation. Electrical stimulation of the FN evokes cardiorespiratory responses via activation of local neurons rather than fibers of passage (36). Therefore, in the present study, we did not address this issue again.

In summary, we found that hyperventilation and pressor responses induced by FN stimulation and the cardiorespiratory responses to hypercapnia were significantly attenuated after bilateral microinjection of IA into the VN\textsubscript{M}. In comparison, the animals’ baseline respiratory variables and cardiorespiratory responses to acute hypoxia were not significantly altered by these lesions. These results suggest that the VN\textsubscript{M} is required for full expression of cardiorespiratory responses to stimulation of the FN as well as to systemic hypercapnia. However, VN\textsubscript{M} neurons appear to be not critical for maintaining eucapnic breathing and cardiorespiratory responses to hypoxia.

\section*{GRANTS}

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