No evidence for long-term facilitation after episodic hypoxia in spontaneously breathing, anesthetized rats

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Janssen, Patrick L., and Ralph F. Fregosi. No evidence for long-term facilitation after episodic hypoxia in spontaneously breathing, anesthetized rats. J Appl Physiol 89: 1345–1351, 2000.—Repeated electrical or hypoxic stimulation of peripheral chemoreceptors has been shown to cause a persistent poststimulus increase in respiratory motoneuron activity, termed long-term facilitation (LTF). LTF after episodic hypoxia has been demonstrated most consistently in anesthetized, vagotomized, paralyzed, artificially ventilated rats. Evidence for LTF in spontaneously breathing animals and humans after episodic hypoxia is equivocal and may have been influenced by the awake state of the subjects in these studies. The present study was designed to test the hypothesis that LTF is evoked in respiratory-related tongue muscle and inspiratory pump muscle activities after episodic hypoxia in 10 spontaneously breathing, anesthetized, vagotomized rats. The animals were exposed to three (5-min) episodes of isocapnic hypoxia, separated by 5 min of hyperoxia (50% inspired oxygen). Genioglossus, hyoglossus, and inspiratory intercostal EMG activities, along with respiratory-related tongue movements and esophageal pressure, were recorded before, during, and for 60 min after the end of episodic isocapnic hypoxia. We found no evidence for LTF in tongue muscle (genioglossus, hyoglossus) or inspiratory pump muscle (inspiratory intercostal) activities after episodic hypoxia. Rather, the primary poststimulus effect of episodic hypoxia was diminished respiratory frequency, which contributed to a reduction in ventilatory drive.

METHODS

Surgical Procedures

The following procedures were performed in 10 male Sprague-Dawley rats (345–425 g). All procedures adhered with the guidelines established by the Institutional Animal Care and Use Committee at the University of Arizona. The animals were anesthetized with a series of intraperitoneal injections of urethane to achieve a dose of 1.3 g/kg. At the end of the experiment, the animals were euthanized with an overdose of urethane administered intravenously. The animals were considered to be in a state of surgical anesthesia if breathing animals is equivocal. In awake dogs (3) and goats (15), episodic hypoxia has been shown to evoke LTF of ventilation, but by different mechanisms. In dogs the response to episodic hypoxia is a persistent increase in tidal volume (3), whereas in goats LTF was shown to be a poststimulus increase in respiratory frequency (15). On the other hand, a recent study in awake humans found no evidence for LTF after episodic hypoxia (12). The reason that hypoxia-induced LTF is a consistent finding in anesthetized rats, and a variable finding in awake animals, could be explained by the difference in the experimental preparations (paralyzed and artificially ventilated vs. spontaneous breathing). LTF has been demonstrated in spontaneously breathing, anesthetized cats after repeated electrical stimulation of carotid chemoreceptors (11), but the influence of hypoxia was not examined.

The purpose of the present investigation was to test the hypothesis that episodic isocapnic hypoxia evokes LTF of pharyngeal airway muscle [genioglossus (GG) and hyoglossus (HG)] and ventilatory pump muscle [inspiratory intercostal (IIC)] activities in spontaneously breathing, anesthetized rats. This study also tested for the first time whether episodic isocapnic hypoxia evokes LTF of a tongue retractor muscle, the HG, which has been shown to be important for regulation of pharyngeal airflow in the rat (6). The stimulation protocol used in the present experiments was modeled after previous studies in artificially ventilated rats, which demonstrated LTF after repeated isocapnic hypoxia (1, 7). The present hypothesis was not supported, and potential reasons for this are discussed.

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they were not responsive to deep pressure application to the paws. Body temperature was monitored with a rectal thermometer (Yellow Springs Instruments) and maintained at 37°C with the use of a servo-controlled heating lamp. Polyethylene catheters (PE-50 tubing) were placed in a femoral vein for administration of fluids and in a carotid artery. The arterial catheter was connected via a three-way stopcock to a pressure transducer (model P23XL, Gould) for measurement of arterial blood pressure and to a heparinized 1-ml syringe for arterial blood sampling. Arterial blood samples were withdrawn in 0.2- to 0.4-ml aliquots over a period of ~10 s and were analyzed within 1 min after sampling for blood gases and pH, by using an Instrumentation Laboratories (model 1640) analyzer. If a base deficit existed, it was corrected by infusion of sodium bicarbonate intravenously. Donor blood was obtained from a urethane-anesthetized littermate and was administered intravenously to replace the volume of blood extracted during arterial sampling. The trachea was cannulated for delivery of inspired gases. Mixtures of O2, N2, and CO2 were delivered to the spontaneously breathing animal by connecting the outflow port of a rotometer to the tracheal cannula with a “t-tube” system. The concentrations of O2 and CO2 in the inspired air were monitored with a very-low-dead-space analyzer (model 1265, Novametrix) placed in series between the tracheal cannula and the t tube. All animals were vagotomized bilaterally at midcervical level during the preparatory surgery.

**EMG recordings.** Electromyogram (EMG) activities of the GG, HG, and IIC muscles were recorded by inserting two fine-wire (diameter = 0.125 mm; Formvar, California Fine Wire) electrodes into each muscle. The wires were insulated except for the terminal 2 mm. Correct electrode placement for the GG and HG muscles was confirmed before each experiment by stimulating the muscles with supramaximal shocks that have been reported during sustained chemoreceptor stimulation in these tracheostomized animals, we acknowledge that changes in pulmonary resistance may alter this relationship. We have shown in recent experiments in urethane-anesthetized rats that changes in peak Pes correlate significantly with both peak IIC and diaphragm EMG activities when breathing is stimulated in hypercapnia (8). These data suggest that Pes is a reliable index of inspiratory pump muscle output in the rat. The product, peak Pes × respiratory frequency (minute Pes), was used to estimate total ventilatory drive. A recent study in our laboratory in anesthetized, spontaneously breathing rats (8) showed that the pattern of change in minute Pes during sustained hypoxia was qualitatively similar to changes in minute ventilation that have been reported during sustained chemoreceptor stimulation in humans and other mammals (16, 18). These data suggest that minute Pes is a useful method for representing total ventilatory drive in the rat.

**Measurement of inspiratory pump muscle function.** Esophageal pressure (Pes) was measured by a saline-filled catheter (PE-160) connected to a pressure transducer (model P23XL, Gould). The tip of the catheter was advanced through the esophagus to the level of the heart. Peak Pes (negative change in pressure during inspiration) was used as an index of tidal volume. Although it is reasonable to assume that tidal volume increases as Pes increases during chemoreceptor stimulation in these tracheostomized animals, we acknowledge that changes in pulmonary resistance may alter this relationship. We have shown in recent experiments in urethane-anesthetized rats that changes in peak Pes correlate significantly with both peak IIC and diaphragm EMG activities when breathing is stimulated in hypercapnia (8). These data suggest that Pes is a reliable index of inspiratory pump muscle output in the rat. The product, peak Pes × respiratory frequency (minute Pes), was used to estimate total ventilatory drive. A recent study in our laboratory in anesthetized, spontaneously breathing rats (8) showed that the pattern of change in minute Pes during sustained hypoxia was qualitatively similar to changes in minute ventilation that have been reported during sustained chemoreceptor stimulation in humans and other mammals (16, 18). These data suggest that minute Pes is a useful method for representing total ventilatory drive in the rat.

**Experimental Protocol.** After the surgical procedures, the preparation was allowed to stabilize for 30 min. Baseline conditions were established by adjusting the inspired gas mixture to 50% O2-balance N2. Phasic GG, HG, and IIC EMG activities were consistently recorded in control conditions. The following procedures were based on protocols outlined in previous LTF studies in anesthetized rats (1, 7).

**Episodic isocapnic hypoxia.** The protocol consisted of 3 (5-min) bouts of isocapnic hypoxia, separated by 5 min of hyperoxia (50% inspired O2). Before the start of the first hypoxic episode, and during a period of stable breathing, arterial blood was sampled to obtain baseline levels of arterial PaO2 (PaO2), arterial PaCO2 (PaCO2), and arterial pH (pHa). Hypoxia was induced by lowering the inspired O2 concentration to 12–13%. Our goal was to produce PaO2 levels of 40–45 Torr during each hypoxic challenge. Isocapnia was maintained by adding 3–3.5% CO2 to the hypoxic inspire. Arterial blood was sampled at the beginning of the third minute of each hypoxic challenge for analysis of PaO2, PaCO2, and pHa. During the 5-min period between each hypoxic challenge, an equal volume of donor blood was administered intravenously to replace the blood lost during arterial sampling.

**Sixty-minute hyperoxic recovery.** After the third (final) hypoxic challenge, the animal was returned to 50% O2 and monitored for 60 min. Arterial blood was sampled at regular intervals during the poststimulation period, and PaCO2 was...
monitored closely. In 4 of 10 rats, it was necessary to add 1–3% CO₂ to the inspired gas mixture during the poststimulation period to correct slight arterial hypoxemia. After each blood sample, an equal volume of donor blood was administered intravenously. Sodium bicarbonate was administered as needed to maintain stable acid-base status.

Response to hyperoxic hypercapnia. After the 60-min poststimulation period, the animal was exposed to 10% CO₂ to obtain maximal levels of activity for each experimental parameter. This response was designated as the “maximal” response to chemoreceptor stimulation. EMG, tongue force, and Pes data that were collected earlier in the experiment were expressed as a percentage of this response.

Determination of apneic threshold. At the end of the protocol, 6 of 10 animals were artificially ventilated (using 50% inspired blood-gas values associated with the apneic threshold). When apnea was achieved, the preparation was allowed to stabilize on artificial ventilation for 5 min. The animal was then removed from the ventilator, and an arterial blood sample was drawn at the reappearance of the first spontaneous breaths to obtain blood-gas values associated with the apneic threshold.

Acquisition and Analysis of Data

Experimental parameters were monitored on a digital storage oscilloscope and/or Grass polygraph. The signals were recorded simultaneously on videocassette recorder tape by using a pulse code modulation system (Vetter) for subsequent off-line analysis.

Peak levels of phasic GG, HG, and IIC iEMG activities, tongue force, and Pes, as well as respiratory frequency, minute Pes, and mean arterial blood pressure (MABP) were measured and averaged over 10 breaths for each data point. EMG, tongue force, and Pes data were expressed as a percentage of their respective maximal responses to 10% inspired CO₂.

Statistical analysis. Changes in peak GG, HG, and IIC iEMG activities, tongue force, and Pes before, during, and after episodic isocapnic hypoxia were analyzed by two-way repeated-measures ANOVA. Changes in respiratory frequency and minute Pes were analyzed with a one-way ANOVA test. Multiple-comparison procedures were performed by using the Student-Newman-Keuls method. A P value of 0.05 or lower was considered significant for all tests.

RESULTS

Episodic Isocapnic Hypoxia

Figure 1 shows GG, HG, and IIC iEMG activities before, during, and after episodic isocapnic hypoxia in one rat. Note that peak iEMG activities are increased to near-maximal levels during each hypoxic challenge, but during the poststimulation period they were not different from baseline levels. Figure 2 shows three (1-min) bins of EMG recordings taken 1 min before, during, and 1 min after the end of the first episode of hypoxia in one rat. Note the reduced EMG burst frequency during the posthypoxia period. Reduced respiratory frequency was consistently recorded after individual episodes of hypoxia in the present study. In this example, there is a slight elevation in iEMG amplitudes after hypoxia. However, this was not a consistent finding in all rats. Increases in EMG activities, when present, consistently returned to baseline levels within 5 min after the end of each hypoxic episode. This finding is reemphasized in the averaged data shown in Fig. 3.

Figure 3 shows average peak IIC, HG, and GG iEMG activities, tongue force, and Pes responses to episodic isocapnic hypoxia. Tongue force and Pes recordings represent the mechanical consequences of respiratory-related neural drive to tongue and inspiratory pump muscles, respectively. The similar patterns of activation of EMG activities and corresponding force and pressure responses suggest that this was a reasonable assumption. Respiratory-related tongue movements were associated with retractive force recordings during all control and stimulation periods, as shown in previous studies in our laboratory (6). The reason that the fraction of maximal tongue force responses were consistently less than EMG activities during control, stimulation, and recovery periods (Fig. 3A) was likely due to coactivation of protrudor (GG) and retractor (HG) tongue muscles and hence cocontraction of these antagonistic muscles. Coactivation of GG and HG muscles has been shown previously to yield a net retraction of the tongue in the rat, whereas denervation of the GG and individual HG activation causes increased tongue retraction force (6). Figure 3 shows that each bout of isocapnic hypoxia caused a significant increase in drive to, and mechanical output by, tongue and inspiratory pump muscles. However, no significant increases in activities were recorded during the 60-min poststimulation period; i.e., there was no evidence for LTF in any of the recordings after episodic hypoxia.

Figure 4 shows average respiratory frequency and minute Pes before, during, and after episodic isocapnic
hypoxia. Each episode of hypoxia caused significant increases in respiratory frequency and minute Pes. Respiratory frequency was reduced relative to baseline levels during the time between each hypoxic event and for a period of 10 min after the end of episodic hypoxia. Consequently, minute Pes was lower than baseline levels between each hypoxic episode and remained reduced for 20 min after the end of the final hypoxic challenge.

Average blood-gas, pHa, MABP, and heart rate data before, during, and after episodic hypoxia are shown in Table 1. Each hypoxic challenge produced a marked decrease in PaO2 and a significant fall in MABP. After the last episode of hypoxia, MABP stabilized at 6–8 mmHg higher than prestimulation control levels. Each episode of hypoxia caused a small (nonsignificant) increase in heart rate. Heart rate was not different from baseline at any point during the poststimulation period. Although there was a trend for higher PaCO2 during the poststimulation period, no significant differences were recorded for PaCO2 before, during, or after episodic hypoxia. In fact, PaCO2 did not deviate by >1.2 Torr from control at any point in the posthypoxic measurement period.

Apneic Threshold

After removal of the animal from artificial ventilation, a period of apnea [44.2 ± 11.7 (SE) s] preceded the resumption of spontaneous breathing. Reversal of apnea was consistently associated with simultaneous reappearance of IIC, HG, and GG iEMG activities, tongue force, and Pes. This suggests that tongue muscle and inspiratory intercostal muscles had similar CO2 thresholds for spontaneous activities. The average PaCO2 at the apneic threshold in six rats was 32.2 ± 2.4 (SE) Torr. The corresponding PaCO2 during spontaneous breathing in hyperoxia (50% O2) was 39.8 ± 1.5

DISCUSSION

The present investigation tested for the first time in spontaneously breathing, anesthetized, vagotomized rats the hypothesis that episodic isocapnic hypoxia evokes LTF of upper airway or inspiratory pump muscle activities. We also tested for the first time whether episodic isocapnic hypoxia evokes LTF of a tongue retractor muscle, the HG, which has been shown to be important for regulation of pharyngeal airflow in the rat (6). These hypotheses were not retained, and possible explanations for this are proposed.

Lack of LTF After Episodic Hypoxia in Spontaneously Breathing Rats

The present findings provide no evidence for LTF after episodic hypoxia in the neural drive to, or mechanical output by, either tongue muscles or inspiratory pump muscles in spontaneously breathing, anesthetized rats. HG, GG, and IIC iEMG activities, as well as tongue force and Pes, were not different from baseline values during any point in the 60-min poststimulation period. Accordingly, we found no evidence for LTF of ventilatory drive (i.e., minute Pes) but rather show short-term depression of breathing, due to diminished respiratory frequency, after episodic hypoxia. These results do not agree with previous findings in artificially ventilated anesthetized rats, which have shown that episodic isocapnic hypoxia evokes poststimulus increases in phrenic (1, 7) and hypoglossal (1) nerve activities.
Critique of Methods

The lack of LTF after episodic hypoxia in the present study could have been influenced by methodological factors. First, the intensity of stimulation of carotid chemoreceptors during each hypoxic episode may not have been severe enough, or the duration of stimulation long enough, to elicit LTF. Or, perhaps the number of hypoxic episodes was below the critical number to evoke LTF. In awake goats, LTF was evoked after 10 (3-min) episodes of isocapnic hypoxia, during which $\text{PaO}_2$ values averaged 47 Torr (15). The critical number of hypoxic episodes required to evoke LTF has been shown to be lower in anesthetized rats. In one study (7), LTF of phrenic nerve activity was shown after three (5-min) episodes of isocapnic hypoxia (PaO$_2$ range 534–51 Torr). In another (1), three (5-min) bouts of 10% inspired O$_2$ were sufficient to elicit LTF of phrenic and hypoglossal nerve activities (blood-gas values during hypoxia were not reported in this study). The present experiments used a protocol of three (5-min) episodes of hypoxia, during which $\text{PaO}_2$ values were carefully regulated between 41 to 43 Torr (Table 1). Clearly, the present protocol was consistent with investigations in anesthetized, artificially ventilated rats showing positive evidence for LTF (1, 7).

Hypocapnia during the poststimulation period could also prevent the manifestation of LTF. However, in the present study, $\text{PaCO}_2$ did not deviate by >1.2 Torr from control at any point in the posthypoxic measurement period. In fact, $\text{PaCO}_2$ was consistently higher, due to mild hypoventilation, after episodic hypoxia, which would favor the expression of LTF. Finally, a significant elevation in arterial blood pressure after episodic hypoxia might mask the expression of LTF. In the present study, MABP was 6–8 mmHg higher than control during the poststimulation period. We do not believe that this increase in blood pressure affected our results, because previous studies in rats have shown that changes in blood pressure of <20 mmHg have minimal effects on respiratory activity (17).
**Apneic Threshold**

We propose that the positive findings for LTF shown in anesthetized, paralyzed, and artificially ventilated rats vs. negative findings in anesthetized, but spontaneously breathing, rats could be related to differences in baseline and poststimulus levels of PaCO₂. In the study by Hayashi and colleagues (7), PaCO₂ was maintained at 1–4 mmHg above apneic threshold via artificial ventilation throughout the experiment. Similarly, Bach and Mitchell (1) reported that PaCO₂ was maintained ~3 mmHg above the apneic threshold via artificial ventilation in their experiments. The present experiments in spontaneously breathing anesthetized, vagotomized rats show that PaCO₂ at apneic threshold [32.2 ± 2.4 (SE) mmHg] was similar to values obtained in anesthetized, vagotomized, artificially ventilated rats (32.5 ± 0.8 mmHg) (7). However, the spontaneously breathing animals used in the present study regulated PaCO₂ ~7 mmHg above the apneic threshold in baseline conditions and nearly 9 mmHg above threshold during the poststimulation period. These data are in agreement with results obtained in a recent study in urethane-anesthetized spontaneously breathing rats, which showed that PaCO₂ at eupnea was 9.0 ± 0.4 Torr higher than at apneic threshold (2). Therefore, LTF may have been masked in the present experiments by the higher baseline and poststimulus levels of PaCO₂. This idea is consistent with findings by Eldridge et al. (4) in paralyzed, vagotomized, artificially ventilated cats. They found that the respiratory after-discharge response after carotid chemoreceptor stimulation becomes more saturated as the CO₂ set point is raised progressively above the apneic threshold (4). Similarly, results from a study by Fregosi and Mitchell (5) suggest that the level to which PaCO₂ is raised above the apneic threshold in paralyzed and ventilated preparations may be critical for the manifestation of LTF. They found in paralyzed, vagotomized, artificially ventilated cats that the CO₂ threshold for spontaneous IIC activity was 5 mmHg higher than the threshold for phrenic nerve activity (5). Thus, to obtain baseline levels of activity for both nerves, end-tidal CO₂ was raised ~2 mmHg above the threshold for spontaneous IIC nerve activities, and nearly 7 mmHg above the threshold for phrenic nerve activity. As a likely consequence, LTF was evoked in IIC nerve activity, but not phrenic activity, after repeated electrical stimulation of the carotid sinus nerve (5).

If indeed LTF is dependent on the arbitrary designation of baseline CO₂ levels in artificially ventilated preparations, and masked by eupneic levels of PaCO₂ in spontaneously breathing preparations, the physiological relevance of LTF in normal breathing should be questioned. However, we concede that the presently used anesthetized, spontaneously breathing rat preparation has limitations compared with awake animals. Although LTF of tidal volume has been demonstrated in anesthetized, spontaneously breathing, vagally intact cats, LTF of the GG muscle was demonstrated only after vagotomy in this preparation (11). This is likely the result of vagally mediated pulmonary stretch receptor mechanisms, which provide stronger inhibition of upper airway compared with ventilatory pump muscles (9). Taken together, these findings suggest that the most relevant difference between our findings and those of Bach and Mitchell (1) and Hayashi et al. (7) is the baseline CO₂ at which the episodic hypoxic stimulation was initiated.

**Reduced Ventilatory Drive After Episodic Hypoxia**

The primary effect of episodic isocapnic hypoxia in spontaneously breathing, anesthetized, vagotomized rats was a reduction in respiratory frequency, lasting ~10 min after the end of stimulation, which contributed to a decrease in total ventilatory drive (i.e., diminished minute PEs). Diminished phrenic nerve burst frequency after repeated carotid chemoreceptor stimulation, termed short-term depression, has been demonstrated in artificially ventilated, vagotomized, anesthetized rats (7). Previous studies in awake humans (12), dogs (3), and goats (15) showed no evidence for a reduction in respiratory frequency after episodic hypoxia. Thus the manifestation of a reduction in breathing frequency after episodic hypoxia in spontaneously

### Table 1. Blood-gas, pHₐ, mean arterial blood pressure, and heart rate measurements obtained before, during, and for 60 min after the end of episodic isocapnic hypoxia

<table>
<thead>
<tr>
<th></th>
<th>PaO₂, Torr</th>
<th>PaCO₂, Torr</th>
<th>pHₐ</th>
<th>MABP, mmHg</th>
<th>Heart rate, beats/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>198.2 ± 5.0</td>
<td>39.8 ± 1.5</td>
<td>7.38 ± 0.01</td>
<td>116.8 ± 2.9</td>
<td>433.8 ± 8.2</td>
</tr>
<tr>
<td>Hypoxia I</td>
<td>42.7 ± 1.5</td>
<td>39.7 ± 2.2</td>
<td>7.38 ± 0.02</td>
<td>89.8 ± 5.5</td>
<td>451.5 ± 9.0</td>
</tr>
<tr>
<td>Hypoxia II</td>
<td>41.1 ± 1.4</td>
<td>37.6 ± 2.2</td>
<td>7.37 ± 0.02</td>
<td>94.5 ± 7.4</td>
<td>448.7 ± 9.2</td>
</tr>
<tr>
<td>Hypoxia III</td>
<td>41.4 ± 0.8</td>
<td>38.8 ± 1.6</td>
<td>7.37 ± 0.02</td>
<td>96.7 ± 7.8</td>
<td>451.1 ± 8.7</td>
</tr>
<tr>
<td>R10</td>
<td>180.2 ± 4.0</td>
<td>40.6 ± 1.1</td>
<td>7.38 ± 0.01</td>
<td>122.7 ± 8.4</td>
<td>429.7 ± 9.6</td>
</tr>
<tr>
<td>R20</td>
<td>181.7 ± 4.4</td>
<td>41.0 ± 1.6</td>
<td>7.37 ± 0.01</td>
<td>126.7 ± 3.3</td>
<td>427.7 ± 8.6</td>
</tr>
<tr>
<td>R30</td>
<td>179.3 ± 3.6</td>
<td>41.0 ± 1.9</td>
<td>7.36 ± 0.02</td>
<td>123.1 ± 4.9</td>
<td>430.9 ± 7.9</td>
</tr>
<tr>
<td>R40</td>
<td>178.7 ± 4.6</td>
<td>41.0 ± 1.6</td>
<td>7.36 ± 0.01</td>
<td>124.6 ± 7.6</td>
<td>434.6 ± 8.5</td>
</tr>
<tr>
<td>R50</td>
<td>173.8 ± 3.8</td>
<td>40.3 ± 1.6</td>
<td>7.36 ± 0.02</td>
<td>124.9 ± 5.3</td>
<td>433.8 ± 8.3</td>
</tr>
<tr>
<td>R60</td>
<td>173.5 ± 5.2</td>
<td>41.0 ± 1.4</td>
<td>7.38 ± 0.01</td>
<td>122.0 ± 4.7</td>
<td>434.6 ± 8.4</td>
</tr>
</tbody>
</table>

Values are means ± SE. PaO₂, arterial PO₂; PaCO₂, arterial PCO₂; pHₐ, arterial pH; MABP, mean arterial blood pressure; Hypoxia I–III, 3 (5-min) episodes of isocapnic hypoxia; R10–60, measurements obtained at 10-min intervals during the posthypoxic recovery period. *Significantly different from corresponding control values, P < 0.05.
breathing animals may be species-dependent. In regard to potential mechanisms, there is evidence that hypoxia-induced depression of respiratory frequency may be due to active inhibition of neurons in the rostral pons or caudal mesencephalon rather than a widespread depression of the central neural activity (10).

Conclusions

The present experiments provide no evidence for LTF of tongue muscle or inspiratory pump muscle activities after episodic isocapnic hypoxia in spontaneously breathing, anesthetized, vagotomized rats. Rather, the only consistent response after episodic hypoxia was a short-term reduction in respiratory frequency contributing to diminished ventilatory drive. Evidence for LTF after episodic hypoxia in awake humans (12), dogs (3), and goats (15) is equivocal. Thus, whether LTF is a consistent response in spontaneously breathing animals remains controversial. Clearly, more studies are needed to resolve the issues that impair a complete understanding of the nature and physiologic relevance of LTF of ventilatory activity.

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