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Hypoxia induced short-term potentiation of respiratory-modulated facial motor output in the rat

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Abstract

Respiratory-modulated facial (VII) nerve discharge includes pre-inspiratory (Pre-I) and inspiratory (I) components. Tonic VII bursting is also present across the respiratory cycle. We tested the hypothesis that hypoxia-induced plasticity of VII motor activity is differentially expressed in Pre-I, I and tonic bursting. Phrenic and VII neurograms were recorded in urethane-anesthetized, vagotomized and ventilated adult rats. A 3 minute isocapnic hypoxic challenge ($\text{PaO}_2 = 33 \pm 2$ mmHg) was used to evoke respiratory short-term potentiation (STP). Pre-I, I and tonic VII activity increased immediately at the initial stage of hypoxia (i.e. acute response) and then progressively increased as hypoxia was maintained. Following hypoxia, I VII activity remained elevated (i.e. post-hypoxia STP) but both Pre-I and tonic activity immediately returned to baseline values. We conclude that STP following hypoxia is preferentially expressed in I compared to Pre-I and tonic VII activity.

Keywords

Hypoxia; facial; short-term potentiation; pre-inspiratory; inspiratory; tonic

1. Introduction

Activation of the alae nasi muscles, which are innervated by the 7th cranial or facial (VII) nerve, can reduce nasal airway resistance (Strohl et al. 1982). Contraction of these muscles therefore contributes to the maintenance of nasal airway patency, particularly when nasal transmural pressures are increased (Bailey and Fregosi, 2006; Gold et al., 1998; Strohl et al., 1982). Consistent with this idea, there is a linear increase in both alae nasi electromyogram activity and force output when ventilatory demands are increased during cycle exercise (Fuller et al., 1995).

During breathing, VII activity includes both phasic and tonic components (Hwang et al., 1988; Hwang and St John, 1988; Lee et al., 2008; Lee et al., 2007; Onimaru et al., 2006), although this designation may depend on the recording conditions (Bailey et al., 2007). Phasic VII activity can be further categorized as either pre-inspiratory (Pre-I), inspiratory (I), or post-inspiratory (Post-I) based on when bursting occurs in relation to phrenic motor activity (Lee et al., 2008; Lee et al., 2007). Although hypoxia-induced neuroplasticity of I phrenic and hypoglossal motor activity has been extensively investigated (see (Mitchell et

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al., 2001; Mitchell and Johnson, 2003) for review), the potential for plasticity in upper airway Pre-I bursting has received relatively little attention (Lee and Fuller, 2010). Hwang and colleagues have described the VII motor response to hypoxia in cats (Hwang et al., 1988), however, to our knowledge the *in vivo* hypoxic response of VII motor output has not been characterized in the rat. Here we examined Pre-I, I and tonic VII activity during and following hypoxia in an anesthetized, ventilated and vagotomized adult rat preparation. Based on prior work indicating that short term potentiation (STP) (Powell et al., 1998) following hypoxia is not expressed in Pre-I hypoglossal activity (Lee and Fuller, 2010), we hypothesized that STP of Pre-I VII discharge would be reduced or absent as compared to phrenic STP.

2. Methods

2.1. Animal preparation

Nine Sprague-Dawley rats (406 ± 10 g, 116 ± 8 days) purchased from Harlan Inc. (Indianapolis, IN, USA) were used. All experimental protocols were approved by the Institutional Animal Care and Use Committee at the University of Florida. Rats were placed in a closed chamber and anesthetized by isoflurane (3–4% in 100 % O₂). Stable anesthesia (2–3% in 100 % O₂) was then maintained by a nose cone. The trachea was cannulated and rats were ventilated (model 683; Harvard Apparatus, Inc, South Natick, MA, USA) during all subsequent procedures. Both the femoral artery and vein were cannulated to permit arterial blood sampling and pressure measurements, and administration of drugs, respectively. Rats were then converted to urethane anesthesia (1.6 g/kg, i.v., Sigma, St. Louis, MO, USA) and given a paralytic agent (pancuronium bromide, 2.5 mg/kg, i.v., Hospira, Inc., Lake Forest, IL, USA). The vagus nerves were sectioned at the cervical level to prevent entrainment of respiratory rhythm with the ventilator. The end-tidal CO₂ partial pressure (P_{ET}CO₂) was recorded via a Capnogard CO₂ monitor connected to the expired line of the ventilator circuit (Novamatrix Medical Systems, Wallingford, CT, USA). Rectal temperature was maintained at 37.5 ± 1 °C by a servo-controlled heating pad (model TC-1000, CWE Inc., Ardmore, PA, USA).

2.2. Nerve recordings

The general procedures have been described previously (Lee and Fuller, 2010; Lee et al., 2008; Lee et al., 2007; Lee et al., 2009). The phrenic nerve was accessed via a ventral approach; the VII nerve was isolated between the nares and the mandible. Both nerves were cut distally and electrical activity was recorded using monopolar silver electrodes. Signals were amplified (1000 x, Model 1700, A-M Systems, Carlsborg, WA, USA), band-pass filtered (0.3 – 10 KHz), and integrated (time constant 100 ms; model MA-1000; CWE Inc., Ardmore, PA, USA). All data were converted to digital signals by a CED Power 1401 data acquisition interface and recorded on a PC using Spike2 software (Cambridge Electronic Design Limited, Cambridge, England).

2.3. Experimental protocol

The CO₂ threshold for rhythmic phrenic bursting was determined after stable nerve recordings were established under hyperoxic (FIO₂ = 0.50–0.60), normocapnic conditions. The P_{ET}CO₂ was gradually reduced by increasing the ventilator rate until rhythmic phrenic bursting ceased for 2 minutes. The ventilator rate was then gradually reduced until rhythmic phrenic bursting resumed. The P_{ET}CO₂ was then maintained at 2–3 mmHg above this recruitment threshold. Baseline blood gas values were established by drawing an arterial blood sample (0.2 ml) (i-Stat, Heska, Fort Collins, CO, USA) after stable recordings were obtained. Rats were then exposed to a three minute bout of hypoxia (FIO₂ = 14.8 ± 0.1), and an arterial blood sample was obtained during the last minute of hypoxia.

2.4. Data analyses

All data were analyzed using Spike 2 software. The criteria used to define the respiratory cycle and Pre-I vs. I burst amplitude were previously described (Lee and Fuller, 2010; Lee et al., 2009). Changes in the peak amplitude of the integrated nerve signals were expressed relative to the maximum value ($\Delta\%$ max) recorded during hypoxia (i.e. $[\text{response} - \text{baseline}]/\text{maximum}$) and during the baseline period ($\Delta\%$ baseline; $[\text{response} - \text{baseline}]/\text{baseline}$). During hypoxia, the calculation of Pre-I and I VII integrated burst amplitude was taken from the “new” baseline associated with increased tonic activity. Tonic VII activity was expressed as arbitrary units (a.u.). Data were averaged over 30 second periods immediately prior to the initial blood sample (baseline), during both the initial and final minute of hypoxia, and at 3 minutes post-hypoxia. A two-way repeated measures analyses of variance (ANOVA) was used to compare phasic phrenic and VII activity across different time points. Tonic VII activity, respiratory pattern and cardiovascular parameters were analyzed using a one-way repeated measure ANOVA. A paired t-test was used to compare blood gas parameters during baseline and hypoxia. Data are expressed as the mean $\pm$ SE. Differences were considered statistically significant when $P < 0.05$.

3. Results

3.1 Arterial blood gases and cardiovascular responses

PaO_2 was reduced from 211 ± 9 mmHg during baseline to 33 ± 2 mmHg during hypoxia as expected ($P < 0.01$). PaCO_2 was similar before and during hypoxia indicating isocapnic hypoxia was induced in the present study (40 ± 2 vs. 40 ± 2 , $P = 0.989$). Hypoxia also induced a significant reduction in mean arterial blood pressure (103 ± 6 vs. 90 ± 8 mmHg, $P < 0.05$) and an increase in heart rate (436 ± 13 vs. 472 ± 8 beats/minute, $P < 0.01$) (see Fig 1).

3.2 Overall respiratory pattern and phrenic nerve activity

The alterations in the respiratory pattern during and following hypoxia are summarized in Table 1. Briefly, the inspiratory duration was reduced during hypoxia ($P < 0.01$, Table 1) and then returned to baseline values at 3 minutes post-hypoxia. The expiratory duration was reduced during the first 30 seconds of hypoxia. However, expiratory duration was increased by the end of the hypoxia exposure ($P < 0.01$), and remained greater than baseline at 3 minutes following hypoxia ($P < 0.05$, Table 1). The effects of hypoxia on phrenic nerve activity were similar with our previous reports (Lee and Fuller, 2010; Lee et al., 2009), and are demonstrated in Fig 1. Specifically, the peak integrated phrenic activity increased by $43 \pm 3 \Delta\%$ max ($79 \pm 9 \Delta\%$ baseline) by the end of the hypoxic episode. The appearance of the integrated phrenic neurogram changed from an augmenting pattern at baseline (Fig. 1Bi) to a decrementing pattern during hypoxia (Fig. 1Biii). This observation may indicate a transition towards a gasping response at the end of the hypoxic exposure (Paton et al., 2006). Phrenic STP was evident following hypoxia as burst amplitude remained elevated at 3 minutes post-hypoxia ($17 \pm 3 \Delta\%$ max; $33 \pm 7 \Delta\%$ baseline) (both $P < 0.01$ vs. baseline, Fig. 2A).

3.3 Facial nerve activity

The VII nerve exhibited both phasic and tonic activity during the baseline period (Fig. 1). Pre-I bursting during the baseline condition was not consistent across experiments. More specifically, the phasic VII burst had a small but detectable Pre-I component during baseline in 56% (5/9) rats (Fig. 1Bi). On average, phasic Pre-I VII bursting was initiated 20 ± 22 ms prior to the phrenic burst. The relatively small Pre-I burst during baseline is consistent with prior reports (Hwang et al., 1988). Conversely, robust Pre-I VII bursting was observed

during hypoxia and the VII-phrenic burst onset difference reached 158 ± 35 ms (Fig. 1Bii–Biii). Following hypoxia, there was some suggestion of inhibition of Pre-I VII bursting. The relative percentage of rats with a clear Pre-I VII burst was similar to the baseline condition (56% or 5/9), but the average VII-phrenic burst onset difference was reduced to 7 ± 7 ms.

Differences were observed in the relative magnitude of Pre-I compared to I VII burst amplitudes during and following hypoxia (Fig. 2A). Pre-I VII burst amplitude was robustly elevated throughout the hypoxic exposure (Fig. 2A; $P < 0.01$) and returned to baseline by 3 min post-hypoxia (-4 ± 4 $\Delta\%$ max, $P=0.422$; 7 ± 19 $\Delta\%$ baseline, $P=0.927$). I VII burst amplitude was also increased during hypoxia, and remained above baseline at 3 minutes post-hypoxia (24 ± 6 $\Delta\%$ max; 122 ± 41 $\Delta\%$ baseline) (both $P < 0.01$ vs. baseline, Fig. 2A). Thus, the offset phase of STP (i.e. post-hypoxia STP) was uniquely observed in I, but not Pre-I VII activity.

In addition to phasic VII activity, tonic VII activity was also enhanced during hypoxia (Fig. 2B, $P < 0.05$). However, tonic activity did not show post-hypoxia STP and was therefore similar to the baseline at 3 minute after the hypoxic exposure (Fig. 2B, $P = 0.819$).

4. Discussion

This study provides the first comprehensive analyses of respiratory-modulated VII motor output during and following hypoxia in an *in vivo* adult rat preparation. The primary finding is that hypoxia induces robust increases in both Pre-I and I VII activity, but STP following hypoxia is only evoked in I VII bursting. Tonic VII bursting also increased substantially during hypoxia but failed to show STP after termination of hypoxia. These findings suggested that hypoxia-induced STP is differentially regulated across Pre-I, I and tonic VII motor outputs.

4.1 Regulation of Pre-I and I VII motor output

Based on bursting patterns relative to I phrenic output, Hwang and colleagues categorized VII motoneurons as Pre-I, I, expiratory (E), inspiratory-expiratory (I-E), expiratory-inspiratory (E-I), and tonic (Hwang and St John, 1988; Lee et al., 2008; Lee et al., 2007). The relative contribution of intrinsic neuronal properties (e.g. rheobase, membrane resistance) vs. synaptic inputs to these motoneuron bursting patterns during spontaneous breathing is not known. It is clear, however, that Pre-I and I VII motor outputs can respond differently to peripheral inputs. For example, Pre-I and I bursting can be differentially modulated by positive end-expired pressure (PEEP) (Lee et al., 2007) and pulmonary C-fiber activation (Lee et al., 2008). High levels of PEEP eliminates I VII bursting although rhythmic Pre-I VII activity persists (Lee et al., 2007). There is also evidence that Pre-I and I VII outputs are controlled by distinct pre-synaptic inputs. Following transection between the medulla and caudal pons – thereby separating the pre-Bötzinger complex and the parafacial respiratory group (pFRG) – phasic Pre-I VII activity can be induced in the absence of I activity by application of a μ -opiate agonist (Onimaru et al., 2006). One interpretation of this result is that Pre-I VII bursting may be driven by neurons in the pFRG (Onimaru et al., 2006). However, Fortuna et al. (2008) point out that the tissue block rostral to the transection in the Onimaru et al. (2006) experiments probably contains a large portion of the Bötzinger region. This is significant because expiratory-augmenting (E-AUG) Bötzinger neurons can also show Pre-I burst patterns during hypercapnic hypoxia (Fortuna et al., 2008). Thus, E-AUG Bötzinger neurons could also be hypothesized to drive Pre-I VII bursting (Fortuna et al., 2008), particularly during intense respiratory stimulation.

Here we show differences in hypoxia-induced plasticity between Pre-I and I activity. One possible interpretation is that robust Pre-I VII activity in our preparation requires a hypoxic

stimulus (i.e. the absence of post-hypoxia STP reflects simply the withdrawal of hypoxia-triggered synaptic inputs that are necessary for the Pre-I burst). However, hypoglossal (XII) motor output, which shows robust Pre-I bursting during normoxic conditions, also fails to show Pre-I STP (Lee and Fuller, 2010). We have also noted that post-hypoxia STP occurs in I, but not E-I XII motoneurons (Lee and Fuller, 2010).

The mechanisms underlying the differences between Pre-I and I STP are not clear. Both outputs responded robustly to hypoxia, but increased synaptic activity is not necessarily required for respiratory plasticity (Bocchiaro and Feldman, 2004). One possibility is that inhibitory inputs to VII motoneurons and/or premotor neurons may restrain Pre-I STP following hypoxia. Consistent with this, we noted that Pre-I VII bursting was actually harder to detect following hypoxia (compared to the baseline condition) despite increased I VII bursting (i.e. I STP). Bötzing neurons with an expiratory-decrementing (E-DEC) pattern could be a source of this inhibition since these cells show prolonged bursting after hypoxia, and may also inhibit E-AUG neurons driving Pre-I bursting (Fortuna et al., 2008). In addition to inhibitory synaptic inputs, centrally-mediated hypoxic depression could also be involved. Central hypoxic inhibition is stronger on E than I neurons (Iizuka and Fregosi, 2007; St John and Bianchi, 1985), and expiratory Bötzing neurons may drive Pre-I bursting (Fortuna et al., 2008). Thus, upon termination of hypoxia and rapid removal of peripheral chemoreceptor (i.e. carotid body) excitatory inputs to Pre-I neurons/premotor neurons, a persistent effect of central hypoxia on E neurons could potentially blunt Pre-I STP following hypoxia.

Reconfiguration of the respiratory network during hypoxia could also have influenced our results. Severe hypoxia results in a transformation of the respiratory pattern from eupnea to gasping (Paton et al., 2006; Rybak et al., 2008). During gasping, the phrenic discharge has a characteristic decrementing pattern, and Pre-I XII activity is substantially reduced or eliminated (Paton et al., 2006). In our experiments, the phrenic discharge pattern began to shift towards a decrementing pattern during hypoxia (Fig. 1), but this was associated with enhanced, not attenuated, Pre-IVII activity. While hypoxia exposure in the present experiments did not induce a “standard” gasping pattern (St John and Paton, 2003), we can not exclude the possibility that even the early phases of the transition between eupnea and gasping may be associated with reconfiguration of respiratory networks with a resultant attenuation of Pre-I VII STP.

4.2. Physiological significance

Activation of the alae nasi muscles reduces nasal airway resistance (Strohl et al., 1982), and thus Pre-I VII nerve activity could dilate and/or stiffen the nasal airway before the onset of inspiratory airflow. However, we and others have noted that Pre-I VII activity becomes considerably more robust during conditions of increased respiratory drive (Hwang et al., 1988). Accordingly, the potential contribution of Pre-I VII bursting to airway patency may be most prominent during conditions such as hypoxia or exercise (Fuller et al., 1995; Sullivan et al., 1996). Pre-I activity could also reduce airflow resistance during the active expirations that occur under these conditions (Abraham et al., 2002; Fuller et al., 1995).

The significance of respiratory STP is not precisely known, but this phenomenon has been proposed to increase the stability of respiratory motor output following periods of hypoxia (Eldridge and Millhorn, 1986). STP of I VII activity following hypoxia would be expected to reduce nasal airflow resistance (Strohl et al., 1982). However, post-hypoxia STP was absent in Pre-I VII motor output suggesting upper airway patency was not increased during Pre-I (or late expiratory) phase following hypoxia. The absence of post-hypoxia STP in Pre-I VII activity could, in theory, restrain expiratory airflow after hypoxia and thus help maintain suitable end-expired lung volumes.

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References

- Abraham KA, Feingold H, Fuller DD, Jenkins M, Mateika JH, Fregosi RF. Respiratory-related activation of human abdominal muscles during exercise. *J Physiol.* 2002; 541:653–663. [PubMed: 12042369]
- Bailey EF, Fregosi RF. Modulation of upper airway muscle activities by bronchopulmonary afferents. *J Appl Physiol.* 2006; 101:609–617. [PubMed: 16675615]
- Bailey EF, Fridel KW, Rice AD. Sleep/wake firing patterns of human genioglossus motor units. *J Neurophysiol.* 2007; 98:3284–3291. [PubMed: 17928550]
- Bocchiaro CM, Feldman JL. Synaptic activity-independent persistent plasticity in endogenously active mammalian motoneurons. *Proc Natl Acad Sci U S A.* 2004; 101:4292–4295. [PubMed: 15024116]
- Eldridge FL, Millhorn DE. Oscillation, gating, and memory in the respiratory system. In: Cherniack, NSWJ., editor. *Handbook of Physiology. The Respiratory System. Control of Breathing.* American Physiological Society; Bethesda: 1986. p. 93–114.
- Fortuna MG, West GH, Stornetta RL, Guyenet PG. Botzinger expiratory-augmenting neurons and the parafacial respiratory group. *J Neurosci.* 2008; 28:2506–2515. [PubMed: 18322095]
- Fuller D, Sullivan J, Essif E, Personius K, Fregosi RF. Measurement of the EMG-force relationship in a human upper airway muscle. *J Appl Physiol.* 1995; 79:270–278. [PubMed: 7559231]
- Gold AR, Smith PL, Schwartz AR. Effect of alae nasi activation on maximal nasal inspiratory airflow in humans. *J Appl Physiol.* 1998; 84:2115–2122. [PubMed: 9609807]
- Hwang JC, Chien CT, St John WM. Characterization of respiratory-related activity of the facial nerve. *Respir Physiol.* 1988; 73:175–187. [PubMed: 3420321]
- Hwang JC, St John WM. Respiratory-modulated activities of motor units of the facial nerve. *Respir Physiol.* 1988; 73:189–200. [PubMed: 3420322]
- Iizuka M, Fregosi RF. Influence of hypercapnic acidosis and hypoxia on abdominal expiratory nerve activity in the rat. *Respir Physiol Neurobiol.* 2007; 157:196–205. [PubMed: 17292680]
- Lee KZ, Fuller DD. Preinspiratory and inspiratory hypoglossal motor output during hypoxia-induced plasticity in the rat. *J Appl Physiol.* 2010; 108:1187–1198. [PubMed: 20150564]
- Lee KZ, Fuller DD, Lu JJ, Ku LC, Hwang JC. Pulmonary C-fiber receptor activation abolishes uncoupled facial nerve activity from phrenic bursting during positive end-expired pressure in the rat. *J Appl Physiol.* 2008; 104:119–129. [PubMed: 17916674]
- Lee KZ, Fuller DD, Tung LC, Lu JJ, Ku LC, Hwang JC. Uncoupling of upper airway motor activity from phrenic bursting by positive end-expired pressure in the rat. *J Appl Physiol.* 2007; 102:878–889. [PubMed: 17082369]
- Lee KZ, Reier PJ, Fuller DD. Phrenic motoneuron discharge patterns during hypoxia-induced short-term potentiation in rats. *J Neurophysiol.* 2009; 102:2184–2193. [PubMed: 19657076]
- Mitchell GS, Baker TL, Nanda SA, Fuller DD, Zabka AG, Hodgeman BA, Bavis RW, Mack KJ, Olson EB Jr. Invited review: Intermittent hypoxia and respiratory plasticity. *J Appl Physiol.* 2001; 90:2466–2475. [PubMed: 11356815]
- Mitchell GS, Johnson SM. Neuroplasticity in respiratory motor control. *J Appl Physiol.* 2003; 94:358–374. [PubMed: 12486024]
- Onimaru H, Kumagawa Y, Homma I. Respiration-related rhythmic activity in the rostral medulla of newborn rats. *J Neurophysiol.* 2006; 96:55–61. [PubMed: 16495360]
- Paton JF, Abdala AP, Koizumi H, Smith JC, St-John WM. Respiratory rhythm generation during gasping depends on persistent sodium current. *Nat Neurosci.* 2006; 9:311–313. [PubMed: 16474390]
- Powell FL, Milsom WK, Mitchell GS. Time domains of the hypoxic ventilatory response. *Respir Physiol.* 1998; 112:123–134. [PubMed: 9716296]

- Rybak IA, O'Connor R, Ross A, Shevtsova NA, Nuding SC, Segers LS, Shannon R, Dick TE, Dunin-Barkowski WL, Orem JM, Solomon IC, Morris KF, Lindsey BG. Reconfiguration of the pontomedullary respiratory network: a computational modeling study with coordinated in vivo experiments. *J Neurophysiol.* 2008; 100:1770–1799. [PubMed: 18650310]
- St John WM, Paton JF. Defining eupnea. *Respir Physiol Neurobiol.* 2003; 139:97–103. [PubMed: 14637316]
- St John WM, Bianchi AL. Responses of bulbospinal and laryngeal respiratory neurons to hypercapnia and hypoxia. *J Appl Physiol.* 1985; 59:1201–1207. [PubMed: 4055598]
- Strohl KP, O'Cain CF, Slutsky AS. Alae nasi activation and nasal resistance in healthy subjects. *J Appl Physiol.* 1982; 52:1432–1437. [PubMed: 6809715]
- Sullivan J, Fuller D, Fregosi RF. Control of nasal dilator muscle activities during exercise: role of nasopharyngeal afferents. *J Appl Physiol.* 1996; 80:1520–1527. [PubMed: 8727535]

Fig. 1. Representative example of phrenic (Phr) and facial (VII) motor output during and following hypoxia

Panel A shows arterial blood pressure (BP) and both integrated ($\int$) and raw Phr and VII neurograms. The period of hypoxia is indicated by the solid line. Note the robust increase in tonic VII activity during hypoxia, and the STP of inspiratory burst amplitude after hypoxia in both the Phr and VII neurograms. Panel B depicts expanded time scale traces of the individual “neural breaths” indicated by i–iv in panel A. In this example, VII nerve burst onset (dotted line) occurred before Phr burst onset (solid line) during baseline conditions (Bi). Pre-I VII bursting became more robust during hypoxia (Bii–iii). The VII-Phr burst onset difference was greater during hypoxia (Bii and Biii), but was not detectable after hypoxia (Biv). Thus, STP was not evident in Pre-I bursting following hypoxia.

Fig. 2. Effects of hypoxia on phasic (A) and tonic (B) VII nerve activity

Panel A demonstrates that Pre-I and I $\int$ VII bursting and I $\int$ Phr bursting were increased during hypoxia. However, only I bursting showed STP following hypoxia. Tonic VII activity increased during but not following hypoxia (Panel B). The labels “Onset” and “End” on the X-axis indicate that the data were obtained over the initial and final 30 seconds of hypoxia; “3min” indicates that data were averaged over 2.5–3 min post-hypoxia. Symbols: *: $P < 0.05$ and **: $P < 0.01$ vs. Baseline; $\Phi\Phi$: $P < 0.01$ comparing Pre-I vs. I VII amplitude; ##: $P < 0.01$ comparing I VII vs. Phr amplitude.

Table 1

Respiratory pattern during baseline, hypoxia and 3 min post-hypoxia

	Baseline	Onset	End	3 min
T _I (sec)	0.39 ± 0.02	0.33 ± 0.01 **	0.27 ± 0.02 **	0.36 ± 0.03
T _E (sec)	0.84 ± 0.03	0.56 ± 0.03 *	1.00 ± 0.08 *	2.4 ± 0.6 *
Frequency (burst•min ⁻¹)	49 ± 1	68 ± 2 **	49 ± 2	28 ± 4 **

Values are means ± SE.

* P < 0.05;

** P < 0.01 compared with the baseline value.