EFFECT OF SEVERE HYPOXIA ON PREFRONTAL CORTEX AND MUSCLE OXYGENATION RESPONSES AT REST AND DURING EXHAUSTIVE EXERCISE

Thomas Rupp* and Stéphane Perrey*

Abstract: Near infrared spectroscopy (NIRS) may provide valuable insight into the determinants of exercise performance. We examined the effects of severe hypoxia on cerebral (prefrontal lobe) and muscle (gastrocnemius) oxygenation at rest and during a fatiguing task. After a 15-min rest, 15 healthy subjects (age 25.3 ± 0.9 yr) performed a sustained contraction of the ankle extensors at 40% of maximal voluntary force until exhaustion. The contraction was performed at two different fractions of inspired O_2 fraction (F_{IO2} = 0.21/0.11) in randomized and single-blind fashion. Cerebral and muscle oxy-(HbO_2) deoxy-(HHb) total-hemoglobin (HbTot) and tissue oxygenation index (TOI) were monitored continuously by NIRS. Arterial O_2 saturation (SpO_2) was estimated by pulse oximetry throughout the protocol. Muscle TOI did not differ between normoxia and hypoxia after the 15-min rest, whereas SpO_2 and cerebral TOI significantly dropped (-6.5 ± 0.9% and -3.9 ± 1.0%, respectively, $P<0.05$) in hypoxia. The muscle NIRS changes during exercise were similar in normoxia and hypoxia, whereas the increased cerebral HbTot and HbO_2 near exhaustion were markedly reduced in hypoxia. In conclusion, although F_{IO2} had no significant effect on endurance time, NIRS patterns near exhaustion in hypoxia differed from normoxia.

1. INTRODUCTION

Since its first application 30 years ago,[1] near infrared spectroscopy (NIRS) has been shown to be an effective tool for monitoring non-invasively central and peripheral changes in oxygenation. It thus may provide valuable insight into the determinants of performance during exercise.[2-7]

* Faculty of Sport Sciences, Motor Efficiency & Deficiency Laboratory, Avenue du Pic Saint Loup, 34090 Montpellier, France. thomas.rupp@univ-montp1.fr

P. Liss et al. (eds.), *Oxygen Transport to Tissue XXX*, DOI 10.1007/978-0-387-85998-9_49,

To date, only a small number of papers have simultaneously measured cerebral (Cox) and muscle (Mox) oxygenation during fatiguing exercise under normoxia and/or hypoxia.[2, 5-9] Previous studies[9, 10] demonstrated that cerebral, but not muscle, tissue shows greater deoxygenation during acute hypoxia at rest and some suggested that Cox was more likely than Mox to limit the maximal whole-body exercise capacity.[5]

The purpose of this study was to explore the effects of severe hypoxia on cerebral (prefrontal lobe) and muscle (gastrocnemius) tissue oxygenation at rest and during a local sustained fatiguing task. We hypothesized that (i) cerebral and muscle deoxygenation would be prevalent during fatiguing exercise under hypoxia and (ii) Cox would drop before the failure in motor performance.

2. METHODS

2.1. Subjects

Fifteen healthy right-footed males (mean ± SE: age 25.3 ± 0.9 yr, height 178.9 ± 1.6 cm, body mass 70.7 ± 1.9 kg) volunteered and gave informed written consent to participate in this study. Subjects were physically active (training 4.9 ± 0.9 $h \cdot wk^{-1}$); had no history of cardiovascular, respiratory, musculoskeletal or neurological disorders; and were free of medication. Subjects were requested to refrain from training on the two days prior to testing and to avoid caffeine and alcohol ingestion for the 12 h preceding the tests. The study procedures complied with the Declaration of Helsinki for human experimentation and were approved by the local ethics committee.

2.2. Experimental Protocol

Each subject visited the laboratory on three occasions (familiarization session and testing sessions) separated by 24 to 48 h. In the preliminary session, a full explanation of the experimental protocol and recommendations were given. Subjects were familiarized with the experimental procedures and individually appropriate adjustments to the ankle-extension device were made. In the testing sessions, they performed a sustained isometric contraction of the ankle extensors at 40% of their maximal voluntary isometric contraction (MVIC) up to exhaustion. This procedure was completed while subjects were exposed in a randomized and single-blind fashion to either room air (normoxia, F_{IO2} = 0.21) or an hypoxic gas mixture (hypoxia, F_{IO2} = 0.11) corresponding to an altitude of ~4500 m. Normobaric hypoxic condition (F_{IO2} = 0.11) was simulated by diluting ambient air with nitrogen via a mixing chamber, with the dilution constantly controlled by a PO_2 probe (Alti-Trainer200, Sport and Medical Technology, Geneva, Switzerland).

The subjects were seated on a padded bench connected to a homemade ankle-ergometer. The foot of the strongest leg (right leg in all instances) was fixed by several semi-rigid Velcro bands on a pedal fitted out with a strain gauge (DEC 60, Captels, St Mathieu de Treviers, France). The knee angle was set at 80° compared with complete extension (0°), and the ankle formed a 90° angle with the pedal. The ergometer was connected to a computer for data acquisition at 1000 Hz and subsequent analysis (MP30, Biopac Systems, Santa Barbara, CA, USA). A monitor provided subjects with visual feedback on the ankle force. Baseline values were first collected for 2 minutes while subjects rested in the exercising position on the ergometer. Then, to reach a steady state,

subjects breathed the appropriate gas mixture at rest for 15 min before continuing the experiments. After a standardized warm-up of the calf muscle, they performed MVICs. The force level target (40% of MVIC) was then determined and represented by a horizontal line on a monitor during the exercise. Subjects were instructed to quickly match their force level to the target line as precisely and as long as possible. Strong verbal encouragement was given to each subject during MVICs and exercise. Exhaustion was defined as a drop in produced force under 30% of MVIC for more than 3 consecutive seconds. Exercise was immediately followed by a post-exercise MVIC to assess force loss. Arterial O_2 saturation (SpO_2) was estimated by pulse oximetry (Pulsox 300i, Konica Minolta Sensing Inc., Osaka, Japan) throughout the protocol.

2.3. Prefrontal Cortex and Muscle Oxygenation

NIRS techniques have been described elsewhere.[4, 11] Cox and Mox were measured continuously and simultaneously with two channels of a four-wavelength (775, 810, 850, 905 nm) high temporal resolution NIRS device (NIRO-300, Hamamatsu Photonics, Hamamatsu City, Japan) throughout the experiment. The probes consisted of one emitter and one detector housed in a black holder. The holders were stuck on the skin with double-sided adhesive tape to ensure no change in their relative positions and to minimize the intrusion of extraneous light and the loss of transmitted NIR light from the field of interrogation. A differential optical pathlength factor (DPF) that takes into account light scattering in tissue is often inserted in the modified Lambert-Beer law used to describe optical attenuation. We decided not to use DPF values as they may vary from one wavelength to another, across subjects, and even over time for a given subject and tissue.[12]

To assess Cox, the detection probes were positioned over the left prefrontal cortical area between Fp1 and F3, according to the modified international EEG 10-20 system. The inter-optode distance was 5 cm and the probe holder was covered and maintained with a homemade black Velcro headband. The prefrontal cortex is known to project to pre-motor areas and to be responsible for movement planning and pacing strategies, as well as decision-making.[13-14] It was also recently shown that decreased frontal cortical oxygenation was associated with reduced muscle force-generating capacity.[15]

To determine local Mox profiles, the probes were attached to the skin on the belly of the right gastrocnemius medialis and in parallel with the long axis of the muscle. The distance between the transmitting and receiving optodes was fixed at 4 cm and the probe holder was covered with a black sweatband maintained with an elastic muff net.

Both cerebral and muscle NIRS data were collected with a sampling frequency of 2 Hz. Relative concentration changes ($\Delta\mu mol \cdot cm$) were measured from resting baseline and pre-exercise level of oxy-($\Delta[HbO_2]$), deoxy-($\Delta[HHb]$) and total-hemoglobin ($\Delta[HbTot] = [HbO_2] + [HHb]$). [HbTot] reflects the changes in tissue blood volume within the illuminated area, [HHb] is known to be a reliable estimator of changes in tissue de-oxygenation status, while $[HbO_2]$ seems to be the most sensitive indicator of regional cerebral blood flow (CBF) modifications.[16] Finally, we used a multidistance spatially resolved tissue oximeter (NIRO-300) that is able to quantify tissue oxy-hemoglobin saturation directly as a tissue oxygenation index (TOI), which is a surrogate measure for cerebrovenous saturation when applied to the head.[17]

2.4. Statistical Analysis

NIRS values were averaged at different time epochs of the protocol: the last 30 s of a 2-min rest period in normoxia, the last 30 s of a 15-min rest period while breathing the appropriate gas mixture, and the last 30 s before the exercise onset. Total exercise time was considered as 100% and data were averaged every 20% of this total time to obtain mean values at 20, 40, 60, 80 and 100% of the performance duration. F_{IO2} (normoxia-hypoxia) and time effects (pre-post 15-min rest period or 0-20-40-60-80-100% of total exercise time) on each of the dependent variables were analyzed using a two-way ANOVA for repeated measures and the Fisher LSD post-hoc procedure when appropriate. Endurance times in normoxia vs. hypoxia were compared with Student's paired *t*-tests. All parameters are expressed as mean ± SE, and the P-value for significance was established at 0.05.

3. RESULTS

3.1. Rest Data

After the 15-min rest period while breathing the hypoxic gas mixture, SpO_2 dropped significantly (from 97.2 ± 0.3 to 90.7 ± 0.8%), whereas no modification was noticed in normoxic condition (from 97.3 ± 0.3 to 97.5 ± 0.2%).

Cerebral [HbTot] was not affected by rest period in either normoxia or hypoxia. Cerebral [HHb] was significantly increased and [HbO_2] was significantly decreased after the 15-min rest in hypoxia but not in normoxia. Cerebral TOI did not statistically differ between normoxia and hypoxia before the 15-min rest period (74.0 ± 1.7 and 73.9 ± 1.2%, respectively) but diverged after it (74.5 ± 1.7 and 69.5 ± 1.0%, respectively), due to a marked cerebral TOI decrease in hypoxia.

Muscle [HbTot] and [HbO_2] were not affected by the rest period in normoxia or hypoxia. Muscle [HHb] was significantly increased after the 15-min rest in hypoxia but not in normoxia. Although muscle TOI did not statistically differ between normoxia and hypoxia before (64.6 ± 1.2 and 65.6 ± 1.4%, respectively) or after (64.3 ± 1.0 and 63.1 ± 1.3%, respectively) the 15-min rest, muscle TOI was slightly decreased in hypoxia but not in normoxia.

3.2. Exercise Data

F_{IO2} had no significant effect on endurance time (458.4 ± 11.0 and 449.4 ± 12.6 s in normoxia and hypoxia, respectively). MVIC dropped similarly after exercise whatever the F_{IO2} condition (-18.0% in normoxia vs. -16.2% in hypoxia). SpO_2 was significantly lower in hypoxia compared with normoxia both pre- (89.7 ± 1.3 and 97.5 ± 0.3%, respectively) and post-exercise (92.6 ± 1.0 and 97.7 ± 0.6%, respectively) but was not significantly affected by exercise.

As presented in Fig. 1, F_{IO2} had a significant effect on cerebral NIRS changes during exercise. Cerebral Δ[HbTot] and Δ[HbO_2] increased from pre-exercise level to exhaustion in normoxia, whereas they stabilized in the last part of exercise (from 80% of total exercise time) in hypoxia. Concomitantly, cerebral Δ[HHb] decreased from pre-exercise level to the last part of exercise in both normoxia and hypoxia and then

stabilized (from 60 and 80% of total exercise time, respectively). Last, cerebral TOI was significantly higher in normoxia compared with hypoxia throughout exercise. Cerebral TOI increased from onset to the last part of exercise in hypoxia and then tended to drop, whereas this parameter was not significantly affected by exercise in normoxia.

Muscle NIRS changes during exercise were not different in normoxia compared with hypoxia (Fig. 1). Muscle Δ[HbTot] and Δ[HHb] increased throughout exercise while Δ[HbO_2] dropped significantly. After an increase at exercise onset compared with pre-exercise level, muscle TOI dropped significantly until exhaustion.

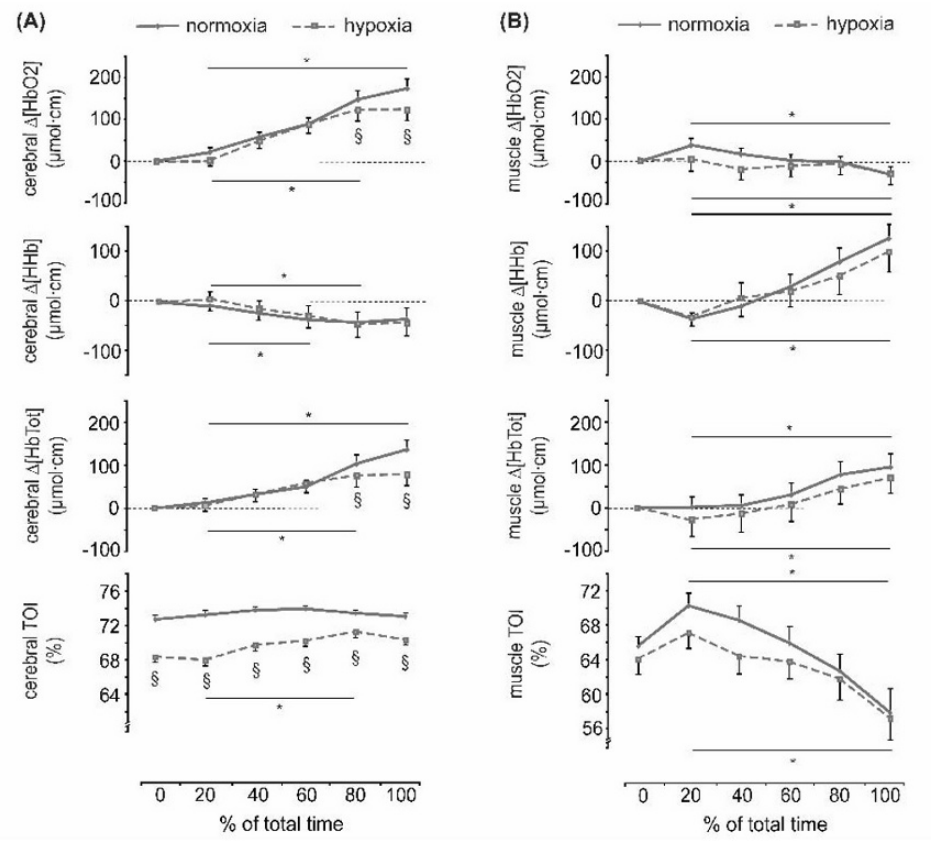

Figure 1. Changes from pre-exercise values in cerebral (A) and muscle (B) oxy-([HbO_2]), deoxy-([HHb]), total-hemoglobin ([HbTot]), and TOI during exercise in normoxia and hypoxia. Values are means ± SE. § different compared with normoxia ($P \leq 0.05$); * different from 20% of total exercise time value ($P \leq 0.05$).

4. DISCUSSION

Local Cox and hemodynamics measured by NIRS reflect cortical activation for various motor tasks[3, 18, 19] while Mox assessed by NIRS is known to reflect the metabolic changes that occur at the muscle site.[4] Our results confirmed recent data[9, 10] and showed that hypoxia during rest and exercise markedly decreased Cox, whereas Mox was not

different between normoxia and hypoxia. Although F_{IO2} had no significant effect on endurance time, NIRS patterns during exercise in hypoxia differed from those in normoxia, as a significant lower cerebral HbTot and HbO_2 was observed near exhaustion, suggesting possible changes in CBF[16]. Finally, Cox patterns might reflect reduced cerebral cortex activity at volitional fatigue in response to dynamic global exercise,[3, 6] but this does not seem to be the case at the end of a local sustained fatiguing exercise.

5. REFERENCES

1. F. F. Jöbsis, Noninvasive, infrared monitoring of cerebral and myocardial oxygen sufficiency and circulatory parameters, *Science* **198**(4323), 1264-1267 (1977).
2. R. A. De Blasi, S. Fantini, M. A. Franceschini, M. Ferrari and E. Gratton, Cerebral and muscle oxygen saturation measurement by frequency-domain near-infra-red spectrometer, *Med. Biol. Eng. Comput.* **33**(2), 228-230 (1995).
3. K. Shibuya and M. Tachi, Oxygenation in the motor cortex during exhaustive pinching exercise, *Respir. Physiol. Neurobiol.* **153**(3), 261-266 (2006).
4. M. Ferrari, L. Mottola and V. Quaresima, Principles, techniques, and limitations of near infrared spectroscopy, *Can. J. Appl. Physiol.* **29**(4), 463-487 (2004).
5. H. B. Nielsen, R. Boushel, P. Madsen and N. H. Secher, Cerebral desaturation during exercise reversed by O2 supplementation, *Am. J. Physiol. Heart Circ. Physiol.* **277**(3), H1045-1052 (1999).
6. A. W. Subudhi, A. C. Dimmen and R. C. Roach, Effects of acute hypoxia on cerebral and muscle oxygenation during incremental exercise, *J. Appl. Physiol.* **103**(1), 177-183 (2007).
7. M. Amann, L. M. Romer, A. W. Subudhi, D. F. Pegelow and J. A. Dempsey, Severity of arterial hypoxemia affects the relative contributions of peripheral muscle fatigue to exercise performance, *J. Physiol.* **581**(Pt1), 389-403 (2007).
8. Y. Bhambhani, R. Malik and S. Mookerjee, Cerebral oxygenation declines at exercise intensities above the respiratory compensation threshold, *Respir. Physiol. Neurobiol.* **156**(2), 196-202 (2007).
9. P. N. Ainslie, A. Barach, C. Murrell, M. Hamlin, J. Hellemans and S. Ogoh, Alterations in cerebral autoregulation and cerebral blood flow velocity during acute hypoxia: rest and exercise, Am J Physiol Heart Circ Physiol **292**(2), H976-983 (2007).
10. J. E. Peltonen, J. M. Kowalchuk, D. H. Paterson, D. S. Delorey, G. R. Dumanoir, R. J. Petrella and J. K. Shoemaker, Cerebral and muscle tissue oxygenation in acute hypoxic ventilatory response test, *Respir. Physiol. Neurobiol.* **155**(1), 71-81 (2007).
11. C. E. Elwell, M. Cope, A. D. Edwards, J. S. Wyatt, D. T. Delpy and E. O. Reynolds, Quantification of adult cerebral hemodynamics by near-infrared spectroscopy, *J. Appl. Physiol.* **77**(6), 2753-2760 (1994).
12. A. Duncan, J. Meek, M. Clemence, C. Elwell, P. Fallon, L. Tyszczuk, M. Cope and D. Delpy, Measurement of cranial optical path length as a function of age using phase resolved near infrared spectroscopy, *Pediatr. Res.* **39**(5), 889-894 (1996).
13. E. K. Miller and J. D. Cohen, An integrative theory of prefrontal cortex function, *Annu. Rev. Neurosci.* **24**,167-202 (2001).
14. M. Suzuki, I. Miyai, T. Ono, I. Oda, I. Konishi, T. Kochiyama and K. Kubota, Prefrontal and premotor cortices are involved in adapting walking and running speed on the treadmill: an optical imaging study, *Neuroimage* **23**(3), 1020-1026 (2004).
15. P. Rasmussen, E. A. Dawson, L. Nybo, J. J. van Lieshout, N. H. Secher and A. Gjedde, Capillary-oxygenation-level-dependent near-infrared spectrometry in frontal lobe of humans, *J. Cereb. Blood Flow Metab.* **27**(5), 1082-1093 (2007).
16. Y. Hoshi, N. Kobayashi and M. Tamura, Interpretation of near-infrared spectroscopy signals: a study with a newly developed perfused rat brain model, *J. Appl. Physiol.* **90**(5), 1657-62 (2001).
17. G. Greisen, Is near-infrared spectroscopy living up to its promises?, *Semin. Fetal. Neonatal. Med.* **11**(6), 498-502 (2006).
18. H. Obrig, C. Hirth, J. G. Junge-Hulsing, C. Doge, T. Wolf, U. Dirnagl and A. Villringer, Cerebral oxygenation changes in response to motor stimulation, *J. Appl. Physiol.* **81**(3), 1174-1183 (1996).
19. L. Mottola, S. Crisostomi, M. Ferrari and V. Quaresima, Relationship between handgrip sustained submaximal exercise and prefrontal cortex oxygenation, *Adv. Exp. Med. Biol.* **578**, 305-309 (2006).