Effects of respiratory muscle training on respiratory CO\textsubscript{2} sensitivity in SCUBA divers.

D.R. PENDERGAST\textsuperscript{1,2}, P. LINDHOLM\textsuperscript{1,2}, J. WYLEGALA\textsuperscript{1,3}, D. WARKANDER\textsuperscript{1}, C.E.G. LUNDEGREN\textsuperscript{1,2}

\textsuperscript{1}Center for Research and Education in Special Environments, \textsuperscript{2}Department of Physiology and Biophysics, \textsuperscript{3}Rehabilitation Sciences, School of Medicine and Biomedical Sciences, University at Buffalo, USA.

Submitted - 1/11/06 - Accepted - 8/1/06

Pendergast DL, Lindholm P, Wylegala J, Warkander D, Lundgren CEG. Effects of respiratory muscle training on respiratory CO\textsubscript{2} sensitivity in scuba divers. Undersea Hyperb Med 2006;33(6):447-453. Typically, ventilation is tightly matched to CO\textsubscript{2} production. However, in some cases CO\textsubscript{2} is retained (SCUBA diving). One factor behind hypoventilation in divers may be low respiratory CO\textsubscript{2} sensitivity. If this is due to inadequate respiratory muscle performance it might be remedied by respiratory muscle training (RMT). We retrospectively investigated respiratory CO\textsubscript{2} sensitivity prior to and after RMT in several groups of SCUBA divers. CO\textsubscript{2} sensitivity (slope of expired ventilation as a function of inspired P\textsubscript{CO\textsubscript{2}}) was measured with a rebreathing technique in 35 subjects with diving experience. RMT consisted of either isocapnic hyperventilation or intermittent vital capacity breaths (twice/minute) against spring loaded breathing valves imposing static and resistive loads generating average inspiratory pressures of \textasciitilde 40 cmH\textsubscript{2}O and expiratory pressures of \textasciitilde 47 cmH\textsubscript{2}O; RMT was performed 30 min/day, 3 or 5 days/week for 4 weeks. Based on pre-RMT CO\textsubscript{2} sensitivity the subjects were divided into three groups: low sensitivity: <2 l/min/mmHg P\textsubscript{CO\textsubscript{2}}, normal: 2-4 l/min/mmHg, and high sensitivity: >4 l/min/mmHg of inspired P\textsubscript{CO\textsubscript{2}}. The normal group had a Pre-RMT CO\textsubscript{2} sensitivity of 2.88±0.60 and a post RMT sensitivity of 2.51±0.88 l/min/mmHg (Mean±SD, n=19, p = n.s). Response in low sensitivity subjects increased from 1.41±0.32 to 2.27±0.53 (n=10, p = 0.002,) while in the high sensitivity group it decreased from 5.41±1.25 to 2.90±0.32 l/min/mmHg (n=6, p = 0.003). These preliminary findings showed that 46% of the subjects had abnormal sensitivity, and suggest that RMT may normalize it in hypo- and hyper-ventilating divers. If the present results are verified, RMT may be an effective means of enhancing safety in CO\textsubscript{2} retaining divers.

INTRODUCTION

Alveolar ventilation is typically tightly regulated to hold Pa\textsubscript{CO\textsubscript{2}} at 40 mmHg in normal healthy subjects (1). However, in certain situations a person may hyper- or hypo-ventilate relative to a given CO\textsubscript{2} production, resulting in a decrease or increase in Pa\textsubscript{CO\textsubscript{2}}, respectively. One situation conducive to CO\textsubscript{2} retention is diving with Self Contained Underwater Breathing Apparatus (SCUBA) which is a common professional, military and sport activity. SCUBA diving involves immersion, exercise, and increased work of breathing due to hydrostatic pressure and airway resistance, all of which expose the respiratory system to increased work of breathing. Voluntary hypoventilation (so called “skipped breathing”), routinely practiced by some divers, may cause hypercapnia. Hypoventilation may also occur due to inability to perform the increased respiratory work required in the physically unique diving environment. Diving-induced CO\textsubscript{2} retention merits investigation because of the potential for CO\textsubscript{2} narcosis (e.g. 2,10) and its tendency to enhance central nervous system oxygen toxicity (CNS-OT) as well as nitrogen narcosis (2, 3). Carbon dioxide retention alone...
may incapacitate a diver due to the narcotic effects of CO₂, a phenomenon originally termed shallow water blackout as observed in oxygen breathing divers when the CO₂ scrubbers in their breathing gear functioned poorly (4). It has also recently been shown in animals that even mild hypercapnia can increase the risk of CNS-OT when hyperoxic gas mixtures are breathed (5). It has furthermore been reported that CO₂ retaining divers may convulse while still within what is generally considered to be the safe limits for hyperoxic exposure (2).

The mechanisms behind CO₂-retention are still unclear, but one factor involved in hypoventilation in divers may be low respiratory CO₂ sensitivity (2, 6, 7). Immersion per se does not alter CO₂ sensitivity in either divers or non-divers (8). Carbon dioxide sensitivity has been reported to be the same at rest and during exercise (9). It has been shown in a few cases that high external breathing resistance may cause CO₂ retention even without prodromal symptoms (no dyspnea) (2,10). It has also been suggested that the ability to tolerate high CO₂ enables working divers to function when breathing becomes cumbersome (11), an “adaptation” with risks since nitrogen narcosis at depth is additive to carbon dioxide narcosis (12), and the combined effects may quickly incapacitate a diver.

It is also possible that part of the hypoventilation in divers is a conditioned behavior learned by divers; Kerem et al (6) showed that divers and ex-divers, who had not been diving for years, hypoventilated compared to a normal population when subjected to an exercise test. However, it was not determined whether this was an adaptation or was due to selection/genetics in the divers. If hypoventilation in divers is due to an acquired behavior it could possibly be normalized by training with a paced breathing pattern.

Another potential cause of hypoventilation in divers is inadequate respiratory muscle performance due to poor function or fatigue which may attenuate the CO₂ response and lead to CO₂ retention. Studies (13) have shown that RMT can improve respiratory muscle function and reduce swimming fatigue.

We hypothesized that respiratory sensitivity to CO₂ quantified with a standard rebreathing test would be normalized by RMT in non-immersed subjects with low CO₂ sensitivity.

This was a retrospective study that analyzed CO₂ sensitivity data collected, and not yet reported, during three previous studies that were specifically designed to determine if RMT improved respiratory muscle and swimming endurance in 35 male certified SCUBA divers (13). Data were pooled from these three studies even though the exact protocol of the RMT varied slightly.

METHODS

The experimental procedure was conducted in conformity with the principles of the Declaration of Helsinki and had been approved by the Institutional Review Board of the University at Buffalo, NY. The subjects gave informed consent to participate.

Thirty-five subjects were recruited from the civilian and military diving communities; they were 26±5 years of age, 178±7 cm tall and weighed 78.9±13.4 kg. The CO₂ sensitivity tests were performed prior to and after two different RMT protocols and a placebo RMT. To check methodological reproducibility, under identical conditions, another 10 subjects (age = 25±1 yrs, ht =181±7cm and wt =84±10 kg) performed respiratory CO₂ sensitivity tests twice, one week apart. These subjects did not participate in any RMT and were studied only to determine the reproducibility of the measurement of CO₂ sensitivity.
Respiratory Muscle Training (RMT)

The RMT consisted of either isocapnic hyperventilation (IHRMT) or breathing against spring-loaded inspiratory and expiratory valves which imposed a combination of static and resistive loading (RMT). Both training protocols were performed for 30 min/day, 3-5 times/week, for 4 weeks. Each subject performed one RMT session per week under the supervision of an investigator. The subjects’ adherence to prescribed RMT schedules, during training at home, was ascertained by weekly reviews of laptop computer recordings of the respiratory pattern used during each training session, as described below. Only subjects that correctly performed all training sessions were included in the data analysis.

Isocapnic Hyperventilation Protocol (IHRMT)

The training device consisted of a plastic bag connected to a mouthpiece equipped with inlet and outlet valves and a rebreathing bag (Fig.1). The device permitted a balance between the volume of rebreathed air from the bag and inhaled fresh air to maintain a constant end-tidal CO₂ (isocapnic) during training. A pressure sensor was connected to the device, and its output was recorded on a laptop computer. The laptop computer also generated “metronome” signals used to pace the breathing frequency and it recorded all breaths in each RMT session. The volume of the bag was initially selected to correspond to approximately 55% of the subject’s measured vital capacity. A suitable breathing frequency (f_b), typically about 30 breaths/minute, was chosen such that it could be maintained for 30 minutes. The combination of bag volume and frequency was aimed at generating 60% of the subject’s maximal voluntary ventilation (MVV) so that f_b = MVV * 0.60 / V_bag.

The next training session began at the highest frequency achieved in the previous session and this was then maintained for 20 min and then followed by an increase of 1-2 breaths/min for the remaining 10 min of the session. When f_b reached 50, the bag volume was increased by 0.1 liter and f_b was reduced to the value that maintained the same level of ventilation, and the cycle was repeated. To ensure that this protocol preserved isocapnia, expired end-tidal CO₂ was monitored once per week using a mass spectrometer (MGA 1100 Medical Gas Analyzer, Perkin-Elmer Corp., Pomona, CA).
Static/Resistive (RRMT) Load Protocol

The subjects assigned to the combination static/resistive load training utilized the same equipment as for IHRMT with minor modifications. Spring loaded expiration and inspiration valves imposing opening pressures of 70.1 ± 16.1 and 61.0 ± 9.6 cmH2O and sustained pressures of 46.6 ± 4.8 and 40.1 ± 3.3 cm H2O were added to the mouthpiece and the rebreathing bag was removed and the connecting tube plugged. A “timer”, displayed on the computer screen, along with an audible beep, was used to prompt each breath inspired and expired against the resistance; this was repeated every 30 seconds. At each beep, the subject took a full inspiration from FRC followed by an exhalation to RV. The subject then removed the mouthpiece, breathed normally, and waited for the next timed cycle. This procedure was repeated for 30 minutes.

CO₂ Sensitivity Test

The CO₂ sensitivity test followed the method described by Read and Leigh (14). Briefly, the subjects re-breathed into and out of a 10 liter recording spirometer filled with a gas mixture of about 8.5% CO₂ and 91.5% O₂. The PCO₂ and PO₂ were monitored continuously at the mouth by mass-spectrometry and the ventilation was derived from the spirometer recording. The PCO₂ and the corresponding ventilation were plotted and the slope calculated as a measure of CO₂ sensitivity (delta \( \dot{V}_E \) versus delta inspired \( \text{PCO}_2 \); l/min Ventilation/mmHg change in \( \text{PCO}_2 \)). Data are presented as mean ± SD for the ratio of expired ventilation to inspired \( \text{PCO}_2 \) (CO₂ sensitivity).

Statistical Analysis

Paired t-tests were used to determine if there were significant effects of RMT on CO₂ sensitivity within each group of subjects. Non-parametric t-tests (Mann-Whitney) were performed between the groups to determine significant differences prior to and after RMT as the groups were subdivided according to high, normal and low CO₂ sensitivity (see below) and therefore were not normally distributed. Statistical significance was accepted with a \( p \leq 0.05 \). In the group of subjects used for testing the reproducibility of the CO₂ sensitivity measurements a linear regression analysis was performed. Sigmastat 3.0, SPSS was used in all tests.

RESULTS

Method

In the group of subjects that did not perform RMT, there was no significant difference between test and retest of CO₂ sensitivity values (paired t-test, \( n=10, p=0.201 \)), and linear regression analysis yielded a significant correlation (\( p=0.001, n=10 \), \( r^2 \) of 0.96, \( y=1.0134x+0.0616 \)) (Fig.2); the slope was not significantly different from 1.0. These data indicate that the CO₂ sensitivity test used for this study was highly reproducible.

![Fig. 2. CO₂ sensitivity (respiratory responses to increased CO₂ in inhaled gas during rebreathing); values are plotted for the 10 subjects from the test-retest reliability study (see methods). This analysis demonstrates that the experience of the first test did not change the sensitivity during the second test, thus this testing was reproducible. A linear regression analysis through the data (solid line) yielded an R² of 0.96, demonstrating the high reliability of the test.](http://archive.rubicon-foundation.org)
Prevalence of CO₂ sensitivity variability

Pre RMT CO₂ sensitivity ranged from 0.9 to 7.2 l/min/mmHg P⁺CO₂ for the divers with an overall mean of 2.9±1.5 l/min/mmHg. The subjects were divided into three groups based on their pre RMT CO₂ sensitivity. Based on reference (7) and our results, the dividing values between the three groups were chosen as follows, low sensitivity: <2 l/min/mmHg, normal: 2-4 l/min/mmHg, and high sensitivity: >4 l/min/mmHg (Fig. 3). The greatest number of subjects (n=19) had a normal CO₂ sensitivity of 2.88 ± 0.60 l/mmHg. There was a relatively high number of subjects (n=10) with low sensitivity 1.44 ± 0.32 l/mmHg, and fewer (n=6) with high CO₂ sensitivity 5.41 ± 1.25 l/mmHg. The group values were significantly different pre RMT (p< 0.001) for the high and low group compared to the normal group.

Effect of RMT on CO₂ sensitivity

The effects of RMT on CO₂ sensitivity are shown in Fig. 3. There was no significant effect of RMT on CO₂ sensitivity in the “normal” group which had a pre-RMT CO₂ sensitivity of 2.88±0.60 and a post RMT sensitivity of 2.51±0.88 (n=19, p = 0.145). However, in the low CO₂ sensitivity group sensitivity increased significantly after RMT from 1.41±0.32 to 2.27±0.53 (n=10, p = 0.002). In the members of the high CO₂ sensitivity group the sensitivity decreased significantly from 5.41±1.25 to 2.90±0.32 (n=6, p = 0.003). Post-RMT values were not significantly different among the three groups (p = 0.44 low vs. normal, p= 0.32 high vs. normal). Accordingly, RMT normalized CO₂ sensitivity in both high and low CO₂ sensitivity groups.

DISCUSSION

The present study indicates that low respiratory CO₂ sensitivity may be normalized by training of the respiratory muscles using paced breathing patterns that specifically enhance respiratory muscle performance as tested by respiratory muscle endurance during isocapnic hyperventilation or by maximal inspiratory and expiratory pressures. Despite years of research, no single explanation has been offered for divers’ CO₂ retention and no test has been shown to reliably explain or predict exertional hypoventilatory hypercapnia (CO₂-retention) in divers, which suggests that this phenomenon could be of multifactorial etiology. Divers’ CO₂ retention is probably influenced by increased hydrostatic pressure across the chest wall (static lung loading), gas density and composition, and by exercise hyperpnea, all of which may cause increased work of breathing. Additionally, lacking capacity of the respiratory muscles to perform
work as well as conditioned behavior may contribute to CO₂ retention.

**Conditioned breathing behavior**

During rest and submaximal exercise in air, V̇E is typically proportional to V̇O₂ and V̇CO₂ while the PaCO₂ is held relatively constant (1). In SCUBA divers however, it has been shown that ventilation may not match CO₂ production resulting in an increase in PaCO₂ (hypoventilation) and consequent CO₂ retention. Remarkably, Young (20) found that excessive respiratory CO₂ sensitivity before navy diving training might be a predictor of failing the diving course. Moreover, it was found that trainees who passed the course exhibited a reduced CO₂ sensitivity at the end of it (20). That diving-related changes in respiratory pattern may be long-lasting is suggested by the findings of Kerem et al (6) who showed that divers and even ex-divers who had not been diving for years hypoventilated compared to a normal population when subjected to an exercise test. Divers who use open circuit systems are constantly encouraged by experience to reduce breathing since every breath changes the divers buoyancy, thus a relatively slow breathing is more comfortable. Also, a novice diver often uses a lot of air, probably due to inexperience with buoyancy technique and a higher stress level and less efficient movements in the water. Although discouraged by instructors, SCUBA divers often compare their air consumption as a means of evaluating diving skill with the resulting motivation to reduce breathing (a.k.a. “skipped breathing”) while at depth.

**Prevalence of low CO₂ sensitivity**

Reduced CO₂ response, hypoventilation, increased end-tidal CO₂ and ensuing hypercapnia have been observed in escape tank instructors (15), trained underwater swimmers (16), non-immersed divers in response to exercise while breathing air (17) as well as O₂ (6). These blunted responses are not universally seen since some studies report that P CO₂ is defended during diving (18,19). The apparent disagreements between studies may be due to individual diver differences or diver training and experience with some divers defending a relatively physiological P CO₂ (at the expense of suffering dyspnea) during actual diving and others allowing the P CO₂ to rise without much discomfort (cf.18). Similar tendencies were observed in the present study of respiratory CO₂ sensitivity with one-half of the subjects defending P CO₂ while the other allowed it to rise prior to RMT. This suggests that the individual differences in respiratory responses to diving may be due to differences in CO₂ sensitivity.

**Carbon Dioxide Sensitivity**

As for an explanation of the effect of RMT to modify the CO₂ sensitivity of the subjects who were either high or low responders it is highly likely that the mechanism was one of changing the effector side of the reflexive respiratory control loop, i.e. the motor response of the respiratory muscles to a given chemoreceptor stimulation. The fact that the CO₂ sensitivity was unchanged by RMT in subjects whose response was normal before RMT is consistent with the notion of a feedback from the effector system to the respiratory
center’s response to rising $P_{\text{CO}_2}$ (21).

CONCLUSION

The present retrospective findings that RMT normalizes respiratory $CO_2$ sensitivity in divers who, before RMT, exhibit an abnormally low or high sensitivity merits future prospective studies in particular to determine if RMT reduces the tendency for $CO_2$ retention during diving in which case this type of training should be employed to protect divers against $CO_2$ intoxication and secondary $O_2$ poisoning. Whether normalization of $CO_2$ sensitivity by RMT in hypersensitive individuals is important in actual diving also remains to be determined. However, given the apparent connection between hypersensitivity and failure to complete demanding dive training, it would be interesting to see if RMT might increase the proportion of candidates passing the training.

ACKNOWLEDGMENT

Support by NAVSEA Grants N61331-03-C-0014 and N773 is gratefully acknowledged.

REFERENCES