

Negative pressure oxygen breathing and head-down tilt increase nitrogen elimination.

J. J. Bodkin¹, T.B. Curry², C. E. G. Lundgren¹

¹Center for Research and Education in Special Environments and the Department of Physiology and Biophysics, University at Buffalo, State University of New York, Buffalo, NY; ²Mayo Foundation for Medical Education and Research, Rochester, MN

Submitted 1/19/06 - Accepted 5/4/06

Bodkin JJ, Curry TB, Lundgren CEG. Negative pressure oxygen breathing and head-down tilt increase nitrogen elimination. *Undersea Hyperb Med* 2006; (33)5:455-462. Negative pressure breathing (NPB) increases the rate of nitrogen elimination, which is thought to be due to an increase in cardiac output due to augmented venous return to the heart. Hyperoxia, however, decreases the rate of nitrogen elimination. The effect of hyperoxia on the increase in nitrogen elimination during NPB is not known. We hypothesized that NPB as and head down tilt (HDT), which is also thought to increase cardiac output, would counteract the detrimental effects of hyperoxia on nitrogen elimination. Nitrogen elimination was measured in 12 subjects while they lay supine breathing 100% O_2 supplied at atmospheric pressure (control), -10 cm H_2O (NPOB₋₁₀), and -15 cm H_2O (NPOB₋₁₅). Nitrogen elimination was also measured in the subjects while they breathed 100% O_2 supplied at atmospheric pressure in the supine position with a 6° HDT. Over a two-hour washout period, NPOB significantly increased nitrogen elimination by more than 14%, although there was no significant difference between NPOB₋₁₀ and NPOB₋₁₅. HDT also significantly increased nitrogen elimination by almost 8%. Neither NPOB nor HDT significantly affected cardiac output but calf blood flow was significantly lower during NPOB₋₁₅. Combining NPB or HDT with 100% oxygen breathing appear to be useful means of increasing nitrogen elimination and should be considered in situations where this effect may be beneficial, such as with oxygen prebreathing prior to decompression.

INTRODUCTION

The physics and physiology of the elimination of inert gases by the human body during decompression or oxygen breathing is complex and not completely understood. It is clear, however, that a major determinant of the rate of transport out of the body of poorly soluble inert gases is the rate of perfusion of the tissues with blood. Factors that increase cardiac output and tissue blood flow, such as exercise, exposure to ambient temperatures above thermoneutral, and immersion in warm water up to the neck, also increase nitrogen elimination during oxygen breathing (1-5). However, oxygen breathing itself may cause a relative decrease in the rate of nitrogen elimination due to its effects on the circulation. Humans breathing a nitrogen-free

gas have lower rates of nitrogen elimination when breathing a hyperoxic gas mixture and higher rates when breathing a hypoxic mixture (6). These changes correspond to changes in cardiac output and calf blood flow under the same conditions.

Previously, we demonstrated that negative pressure breathing of a normoxic, nitrogen-free gas increases the rate of nitrogen elimination compared with breathing the same gas at ambient pressure (7). It has also been shown that pre-breathing oxygen at negative pressure delays and reduces both venous gas embolism and the incidence of decompression sickness after hypobaric decompression compared with pre-breathing oxygen at normal pressure (8). However, neither the

optimal negative pressure for the purpose of augmenting nitrogen elimination nor the effects of hyperoxia on nitrogen elimination during negative pressure breathing have been studied. Oxygen pre-breathing is a standard method of decreasing the risk of decompression sickness during hypobaric decompression and 100% oxygen is also routinely used during decompression after hyperbaric exposures and during treatment of decompression sickness. Therefore, we decided to study the effect of negative pressure breathing with 100% oxygen (negative pressure oxygen breathing, NPOB) on nitrogen elimination.

We tested the hypothesis that nitrogen elimination during 100% oxygen breathing would be increased by negative pressure breathing and adopting a head-down position.

METHODS

Subjects

The study protocol was approved in advance by the Institutional Review Board for Human Subjects in Research of the University at Buffalo, State University of New York. Written informed consent was obtained from all subjects ($N = 12$). No stipulation on gender was made while recruiting subjects however no females volunteered. Subjects were screened for diseases and medication use that could affect pulmonary or cardiovascular function.

Measurements of nitrogen elimination

Subjects underwent measurements of nitrogen elimination while lying supine and breathing 100% oxygen at ambient pressure (control), -10 and -15 cm H_2O negative pressure breathing (NPOB₋₁₀ and NPOB₋₁₅). Nitrogen elimination was also measured while subjects breathed 100% oxygen at ambient pressure in a supine position with a 6° head-down tilt (HDT). The order of studies was determined by subject

recruitment and availability. A minimum of twenty-four hours was required between experiments for each subject to allow the body tissues to re-equilibrate with air. Similarly, the subjects were restricted from diving, flying, or consuming caffeine for twenty-four hours prior to experiments.

Subjects were positioned supine in a sealed polyethylene tent maintained at 25 ± 0.2 °C with a heat exchanger and a fan (Figure 1). The tent was flushed with a nitrogen-free, normoxic gas mixture (79% argon/21% oxygen) to maintain the ambient nitrogen concentration than 1%. The amount of nitrogen eliminated was measured using a rebreathing method and setup used previously in this laboratory (6, 7, 9). Briefly, the subject, wearing a nose clip and a mouth piece, breathed for a specified time through a closed-circuit loop with a carbon dioxide absorber and a bag containing 100% oxygen. The subject then exhaled to RV and began to breathe from a second bag containing fresh 100% oxygen. The bag from which the subject had been rebreathing was then emptied and the volume and temperature of the gas in each rebreathing bag was measured using a gasometer (Precision Scientific, Chicago, IL). Nitrogen content was quantified using a gas chromatograph (Hewlett Packard, Avondale, PA) equipped with a thermal conductivity detector and a molecular sieve column (Alltech, Deerfield, IL). The amount of nitrogen remaining in the lungs and breathing circuit after the subject was switched to a fresh bag was calculated from the residual volume of the subject's lungs (determined separately), the dead space of the circuit, and the nitrogen content of the sample. The residual nitrogen was subtracted from the nitrogen content of the new bag and added to the contents of the sample with which it was associated. In this manner, all of the nitrogen eliminated during a given rebreathing period was included in the results for that sample. The volume of eliminated

nitrogen was expressed at STPD using body temperature (37°C), the temperature of the gas in the rebreathing bags and the gasometer, and the average atmospheric pressure during the experiment. The amount of nitrogen eliminated from the body and collected in the rebreathing bags was measured thirteen times over 125 minutes during each experiment (a 5 min period of rebreathing followed by twelve 10 min periods).

Breathing gas was supplied at negative pressure as previously described (7). Briefly, the bags containing breathing gas were placed in a barrel (outside of the tent) that was kept at ambient pressure (control) or evacuated to -10 or -15 cm H_2O (-1.0 or -1.5 kPa). In separate experiments, A 6° head-down tilt position was obtained by inclining the tent bottom and breathing apparatus while the subject lay supine. All experiments were carried out at atmospheric pressure.

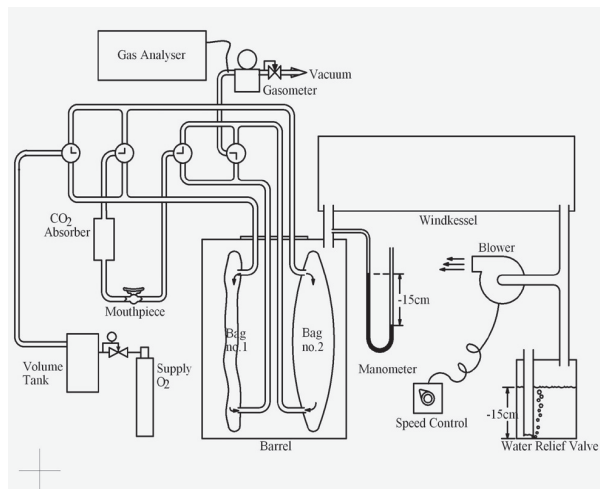


Fig. 1. Experimental setup for supplying oxygen at negative pressure. The barrel holding the bags containing oxygen is connected to a blower. To protect the subject from excessive negative pressure and to minimize fluctuations in barrel and bag pressures during breathing to less than 0.5 cm H_2O (49 Pa), a water relief valve is set to a depth of 10 or 15 cm (the desired negative pressure) and connected to the blower and a windkessel (an air-filled tank used to dampen pressure fluctuations).

Measurements of cardiovascular parameters

Heart rate (HR), stroke volume (SV), cardiac output (CO), blood pressure (BP), and calf blood flow (CBF) were measured during each rebreathing period. Baseline measurements were first made while the subject lay supine in the tent breathing air at atmospheric pressure. CBF was measured with limb plethysmography using a mercury-in-silastic strain gauge (Parks Medical Electronics, Aloha, OR). A BioZ Impedance Cardiograph Monitor (CardioDynamics, San Diego, CA) was used to measure HR, BP, SV, and CO. Cardiac index (CI) was calculated from the CO and body surface area (BSA).

Determination of total body nitrogen

The total body nitrogen content of the subjects was calculated from the solubility coefficients for nitrogen in oil and water and the fat and lean masses of the subject as previously described (7).

Statistical analysis

Friedman non-parametric analysis of variance (ANOVA) was used to analyze for differences in the amount of nitrogen eliminated and cardiovascular parameters between the intervention and control experiments. An alpha error = 0.05 was used for all statistical analysis. We hypothesized that nitrogen elimination would be increased by NPOB and HDT but less than the 40% increase we observed during NPB of a nitrogen-free normoxic gas mixture. A sample size calculation for an ANOVA analysis of 4 groups using a minimal detectable difference in the amount of nitrogen eliminated of 10% and a standard deviation of 5% (based on repeated measurements of nitrogen elimination during breathing at atmospheric pressure) to achieve a power ≥ 0.80 with an alpha error = 0.05 yielded a minimal sample size of 7.

RESULTS

All 12 subjects completed 2-3 control experiments and one experiment with each of the experimental conditions (NPOB₋₁₀, NOPB₋₁₅, and HDT). The average physical characteristics of the subjects are shown in Table 1.

Table 1. Physical characteristic of the subjects (N = 12). Data are mean values \pm SD.

Age	Height (m)	Weight (kg)	Body	
			Fat (%)	BSA (m ²)
26.2 \pm 10.0	1.79 \pm 0.1	82.4 \pm 10.7	16.1 \pm 5.0	2.0 \pm 0.1

Nitrogen elimination

The total volume of nitrogen eliminated during NPOB at both levels of pressure (NPOB₋₁₀ = 667.7 \pm 137.2 mL, NPOB₋₁₅ = 662.1 \pm 159.0 mL) was significantly greater compared with normal pressure breathing (control) (605.3 \pm 145.5 mL) (mean \pm SD). This represents an average increase of 14.5% during NPOB. Significant differences between NPOB and control were seen at all time points during the experiment (Figure 2). There was no

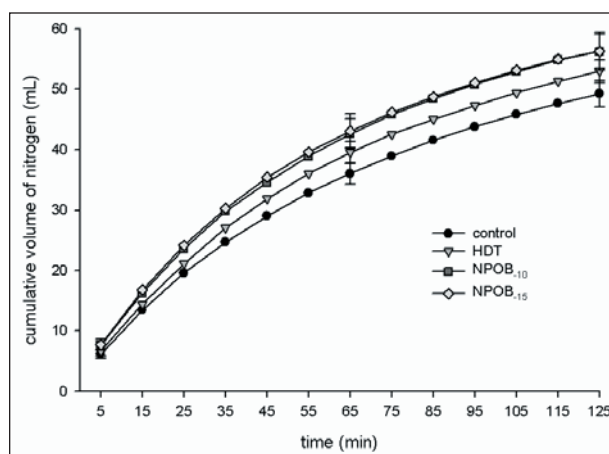


Fig. 2. Nitrogen volumes eliminated during control and negative pressure breathing. Data points are mean values \pm SD (error bars on values other than the 5, 65, and 125 min data points are omitted for clarity). Significant differences between negative pressure breathing (NPOB₋₁₀ and NPOB₋₁₅) and head-down tilt (HDT) compared with control were found at each point in time.

significant difference in the volume of nitrogen eliminated between the two different levels of NPOB at any point during the experiment. Nitrogen elimination during HDT (645 \pm 166.8 mL) was significantly (7.9%) greater compared with control (supine position). On average, the amount of nitrogen eliminated in 125 min under control conditions was eliminated in 88.5 \pm 25.5 min during NPOB₋₁₀, 88.9 \pm 23.7 min during NPOB₋₁₅, and in 103.5 \pm 12.4 min during HDT, although these differences were not significant. An interclass correlation coefficient was calculated for two control measurements in each of the 12 subjects to demonstrate the reliability of the nitrogen elimination measurements (R = 0.997).

Cardiovascular parameters

The baseline cardiovascular parameters were not significantly different between the experimental conditions. Heart rate was significantly lower and SV was significantly higher during HDT and NPOB₋₁₀ than during control. Calf blood flow was significantly lower during NOPB₋₁₅. Blood pressure was not significantly different between any of the study conditions (Table 2).

	control	HDT	NPOB ₋₁₀	NPOB ₋₁₅
HR (bpm)	69.3 \pm 3.2	58.6 \pm 1.4*	60.9 \pm 1.8*	64.0 \pm 1.5
SV (mL)	82.6 \pm 5.0	100.0 \pm 1.3*	97.2 \pm 1.3*	92.8 \pm 2.1
CI (L \cdot min ⁻¹ \cdot m ⁻²)	3.0 \pm 0.2	2.9 \pm 0.1	3.0 \pm 0.1	3.0 \pm 0.1
SBP (mmHg)	124.6 \pm 4.6	121.9 \pm 2.1	121.7 \pm 2.6	124.1 \pm 3.1
DBP (mmHg)	74.4 \pm 7.9	72.2 \pm 2.1	72.8 \pm 3.1	73.6 \pm 2.6
CBF (mL \cdot min ⁻¹ \cdot 100g ⁻¹)	3.37 \pm 1.32	2.36 \pm 0.29	2.39 \pm 0.40	2.06 \pm 0.20*

Table 2. Cardiovascular parameters during nitrogen elimination measurements. Data are mean values \pm SD. HDT = 6° head down tilt, NPOB₋₁₀ = negative pressure oxygen breathing at -10 cmH₂O, NPOB₋₁₅ = negative pressure oxygen breathing at -15 cmH₂O, HR = heart rate, SV = stroke volume, CI = cardiac index, SBP =

systolic blood pressure, DBP = diastolic blood pressure, CBF = calf blood flow. * = significantly different from control.

DISCUSSION

In the current study, negative pressure breathing with 100% oxygen significantly increased the rate of nitrogen elimination by more than 14% compared with breathing gas supplied at atmospheric pressure. The same amount of nitrogen eliminated over 125 min of the control experiments was eliminated in about 35 min less time during negative pressure breathing. Thus, the increase in the rate of nitrogen elimination should be sufficient to impact the amount of time needed for oxygen pre-breathing prior to decompression, decompression after diving, and treatment of decompression sickness. It was also shown that the more modest level of negative pressure (-10 cm H_2O) was just as effective as the greater level (-15 cm H_2O). These results are consistent with the observation that pre-breathing oxygen at negative pressure prior to hypobaric decompression delays the onset and amount of venous gas embolism and reduces the incidence of decompression sickness compared with pre-breathing oxygen at normal pressure (8). Similar, although smaller, increases in nitrogen elimination were seen during HDT.

We previously showed that NPB of a normoxic nitrogen-free gas mixture increased the rate of nitrogen elimination by almost 40% (7). However, this laboratory has also shown that high levels of hyperoxia results in an almost 17% decrease in nitrogen elimination that is accompanied by decreases in CO and CBF, presumably due to reflex vasoconstriction (6). The results of the current study show that hyperoxia reduces the effect of negative pressure breathing on nitrogen elimination but does not eliminate it.

Some authors have found that breathing

spontaneously against negative pressure increases cardiac output (10-13). The increase is thought to be due to an augmentation of venous return (preload) and increase in central blood volume as a result of decreased intrathoracic pressure. However, others have found that cardiac output does not increase during negative pressure breathing (14-16) or that any increase is small, variable, and temporary (17). Negative inspiratory pressure has even been shown to decrease stroke volume and ejection fraction (18) which is likely due to an increase in left ventricular afterload, although the physiology of the cardiovascular effects of negative pressure breathing is complex (19). Similarly, while head-down tilt is classically thought to increase cardiac output (20-22), others have found that it does not (23-31). In the current study, significant but offsetting changes were observed in HR and SV during HDT and $NPOB_{-10}$, and as a result, CO was unchanged by $NOPB$. This may have been due to cardiovascular reflexes stimulated by an increase in central blood volume during negative pressure breathing. As in our previous work, no changes in HR or SV were observed during with negative pressure breathing at -15 cm H_2O . However, since the primary outcome of the study was the amount of nitrogen elimination, it was not powered to detect differences in cardiovascular parameters and there may have been differences in these variables that we could not detect in the current study. However, it is not cardiac output *per se* that determines nitrogen elimination, but rather the perfusion of tissues that contain nitrogen. Balldin and Linér (32) have shown that $NPOB$ causes a dramatic (68%) increase in adipose tissue blood flow compared with normal pressure breathing. Based on the solubility of nitrogen in oil and water, adipose tissue contains approximately five times more dissolved nitrogen than watery tissues (such as muscle) at steady-state. Thus, the reason

for the increase in nitrogen elimination during NPOB may be a redistribution of blood flow to tissues that contain a large amount of dissolved nitrogen. Clearly, however, the mechanisms for the increase in nitrogen elimination during NPB and HDT needs further study.

Experimental error

Since the breathing system was subjected to negative pressure, it was essential to prevent leakage of nitrogen-containing air into the system during the experiment. The extensive leak testing performed to verify the absence of any inward leaks has been previously described (7) and was repeated before, after, and several times during the course of this study. Most of the breathing circuit was inside the tent that was flushed with nitrogen-free (argon/oxygen) gas and leaks in this portion of the rebreathing circuit, including any around the subjects' mouthpiece, would not have contained enough nitrogen to account for the increase in nitrogen elimination during negative pressure breathing. Argon was monitored for. The absence of argon (which would have signified an inward leak from around the mouthpiece or the circuit inside the tent) in the breathing gas during NPOB was confirmed by mass spectrometry of serial samples of the breathing gas.

Impedance cardiography, a non-invasive method to quantify stroke volume and cardiac output, is easily implemented but there is debate as to the accuracy of the absolute values of cardiac output generated through this technique. It is generally agreed that impedance cardiography reliably reflects trends in stroke volume (33) and it has been utilized successfully by this laboratory for this purpose in the past (6, 9, 34). We further confirmed the accuracy of impedance cardiography in our hands by comparing it to a single breath CO_2 technique (35) which has itself been validated against the direct Fick method (36). Twelve subjects underwent measurements of cardiac output

using impedance cardiography and the single breath CO_2 technique, six while sitting and six lying supine, both during normal pressure and negative pressure breathing (data not shown). There were no significant differences between the impedance method and the revised one-step CO_2 rebreathing method (Wilcoxin signed ranks test) and the results obtained by the two methods correlated well ($R = 0.83$).

CONCLUSIONS

The increase in nitrogen elimination rate seen in the current study during NPOB provides mechanistic support for the observation that NPB decreases venous gas emboli and decompression sickness prior to hypobaric decompression compared with 100% oxygen alone. Negative pressure breathing should be alone. Negative pressure breathing should be considered as a means of increasing nitrogen elimination prior to or during decompression and during recompression for the purpose of treating decompression sickness. The exact mechanism of the increase in nitrogen elimination needs further study but it may be due to a redistribution of blood flow to tissues containing large amount of dissolved nitrogen. Negative pressure breathing appears to be a well-tolerated and practical way of accelerating nitrogen elimination compared to other available methods and can be easily and effectively applied, with or without oxygen, using appropriate technical means.

ACKNOWLEDGMENTS

Supported by U.S. Navy Coastal Systems Station award no. N61331-01-C-0017 and the Department of Physiology and Biophysics University at Buffalo, State University of New York, Buffalo, NY and the Department of Anesthesiology, Mayo Clinic, Rochester, MN.

We would like to acknowledge the statistical assistance of Cathy Buyea and the technical assistance of Curtis Senf, Andrew Barth, and Dean Markey.

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