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Exposure to Mild Hyperbaric Oxygen Increases Blood Flow and Resting Energy Expenditure but not Oxidative Stress

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Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

Original Research Article

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ABSTRACT

Aims: This study examined the effects of exposure to mild hyperbaric oxygen on blood flow and resting energy expenditure.
Study Design: Clinical study.
Methods: Fourteen healthy women were exposed to mild hyperbaric conditions at 1.25 atmospheres absolute with 36.0% oxygen for 50 min. Their heart rate, peripheral oxygen saturation, blood flow, resting energy expenditure, derived-reactive oxygen metabolites, and biological antioxidant potential were monitored, and the values before and after exposure were compared.
Results: Heart rate decreased after exposure to mild hyperbaric oxygen. In contrast, peripheral oxygen saturation, blood flow, and resting energy expenditure increased after exposure. There were no changes in the levels of derived-reactive oxygen metabolites or biological antioxidant potential after exposure.
Conclusions: Exposure to mild hyperbaric oxygen increases blood flow and metabolism without increasing levels of oxidative stress.

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Keywords: *Biological antioxidant potential; blood flow; derived-reactive oxygen metabolites; heart rate; mild hyperbaric oxygen; peripheral oxygen saturation; resting energy expenditure.*

1. INTRODUCTION

An elevation in atmospheric pressure accompanied by an increase in oxygen concentration enhances the partial pressure of oxygen and increases the levels of oxygen dissolved in plasma. Hyperbaric conditions at 2–3 atmospheres absolute (ATAs) with an oxygen concentration of 100% are generally used for hyperbaric oxygen therapy [1,2]. Hyperbaric oxygen therapy is used for the treatment of temporary hypoxia [3], tissue repair after burn injury [4], intractable ulcer [5,6], open fractures, and crush injuries [7]. However, the conditions used in hyperbaric oxygen therapy are thought to induce the excessive production of reactive oxygen species in several tissues and organs [8].

In previous studies, we found that mild hyperbaric conditions (1.25 ATAs with 36.0% oxygen) can be used to increase oxidative capacity in cells and tissues [9,10]. Whereas concentrations of oxygen higher than 40% cause side effects such as enhanced levels of oxidative stress [11] and/or increased numbers of invasive inflammatory cells [12], conditions of mild hyperbaric oxygen do not cause enhanced levels of oxidative stress [11]. In addition, we previously observed in animal experiments that type 2 diabetes [13–16], diabetes-induced cataracts [17], hypertension [18], type II collagen-induced arthritis [19], and age-related decline in muscle oxidative enzyme activity [20] were inhibited and/or improved by exposure to mild hyperbaric oxygen.

We estimate that the amount of oxygen dissolved in plasma under mild hyperbaric conditions is 2.76 times greater than that under normal conditions, when the atmospheric pressure is 1.25 times greater and the oxygen concentration is 1.72 times higher than normal. Thus, it is plausible that the increased dissolved oxygen enhances metabolism in cells and tissues. In this study, we examined the effects of exposure to mild hyperbaric oxygen on blood flow and resting energy expenditure. Healthy women were exposed to mild hyperbaric conditions for 50 min. Values for heart rate, peripheral oxygen saturation (SpO_2), blood flow, resting energy expenditure, derived-reactive oxygen metabolites (dROMs), and biological antioxidant potential (BAP) were obtained before and after exposure to mild hyperbaric oxygen and then compared. dROMs and BAP were used as indices of oxidative stress and antioxidant capacity, respectively. Our results show that exposure to mild hyperbaric oxygen increases blood flow and metabolism without increasing levels of oxidative stress.

2. MATERIALS AND METHODS

2.1 Participants and Exposure to Mild Hyperbaric Oxygen

For this study, we used a mild hyperbaric oxygen chamber that we had designed for use in human experiments (Japan Patent No. 5076067, dated September 7, 2012). The chamber consists of an oxygen tank (length, 240 cm; width, 95 cm; height, 95 cm; weight, 140 kg) in which a single participant could lie down and a control box (length, 55 cm; width, 50 cm; height, 120 cm; weight 70 kg) containing an oxygen concentrator and an air compressor. The atmospheric pressure and oxygen concentration were controlled using a computer-

assisted system in the control box. The interior of the mild hyperbaric oxygen chamber was automatically maintained at a temperature of 22±2ºC with a relative humidity of 45–55%.

Using this chamber, 14 healthy women (age, 18.4±0.5 years; height, 1.58±0.03 m; body weight, 48.9±3.3 kg; body mass index, 19.5±0.9 $kg \cdot m^{-2}$; values are means ± standard deviations) were first exposed to normobaric conditions (1.00 ATA with 20.9% oxygen) for 50 min as the control. The same participants were then exposed to mild hyperbaric oxygen for 50 min on a subsequent day.

2.2 Heart Rate and SpO_2

Heart rate (beats/min) and SpO_2 (%) were monitored on the right forefinger of each participant, with the participant in a supine position, using a pulse oximeter (PULSOX–300i; Konica–Minolta Sensing Co., Ltd.; Sakai, Japan) before and after exposure to normobaric conditions or mild hyperbaric oxygen.

2.3 Blood Flow

Blood flow (mL/min/100 g tissue) was measured using a laser Doppler flowmeter (FLO–N1; NeuroScience, Inc.; Tokyo, Japan). The flowmetry probe was attached to the back of the participant's right hand. Blood flow was continuously measured during exposure to normobaric conditions or mild hyperbaric oxygen. The data were transferred to a personal computer via an interface (EFA/400; Distributed Design Concept; Dover, NH, USA) from the flowmeter and analyzed using the data recording software LogWorx, ver. 1.804 (Distributed Design Concept).

2.4 Resting Energy Expenditure

Resting energy expenditure (kcal) was measured using a metabolic analyzer (MedGem indirect calorimeter; Microfile, Medical Home Solutions, Inc.; CO, USA) before and after exposure to normobaric conditions or mild hyperbaric oxygen. The data were transferred to a personal computer and analyzed using the software MedGem Analyzer (Microfile, Medical Home Solutions, Inc.).

2.5 dROMs and BAP

The levels of dROMs and BAP were estimated before and after exposure to normobaric conditions or mild hyperbaric oxygen. Blood samples were obtained from the left forefinger. A free radical and antioxidant potential determination device (Free Radical Analytical System 4; Health & Diagnostics; Grosseto, Italy) was used to measure the levels of dROMs and BAP [21,22]. dROM values indicate serum hydroperoxide levels and are obtained by measuring the amount of N, N-diethyl-p-phenylenediamine oxidized by hydroperoxide-derived free radicals from the plasma sample [23, 24]. dROMs are expressed in Carr unit, named after an Italian biologist who designed a scale based on data from >5000 nonsmoking, healthy participants aged 14–80 years (1 Carr unit = 0.08 mg hydroperoxide/100 mL H_2O_2) [21]. The average level of dROMs for healthy individuals is approximately 300 Carr units [23]. BAP levels are determined based on the capacity of the plasma sample to reduce ferric ions to ferrous ions [24].

2.6 Statistical Analysis

Means and standard deviations were calculated from individual values using standard procedures. The Student's *t*–test was used for statistical comparisons; significance was indicated by a *P*-value <0.05.

3. RESULTS

3.1 Heart Rate

There was no change in heart rate after exposure to normobaric conditions (Fig. 1A). In contrast, the heart rate decreased after exposure to mild hyperbaric oxygen (Fig. 1B).

3.2 SpO_2

There was no change in SpO_2 after exposure to normobaric conditions (Fig. 1C). In contrast, the SpO_2 increased after exposure to mild hyperbaric oxygen (Fig. 1D).

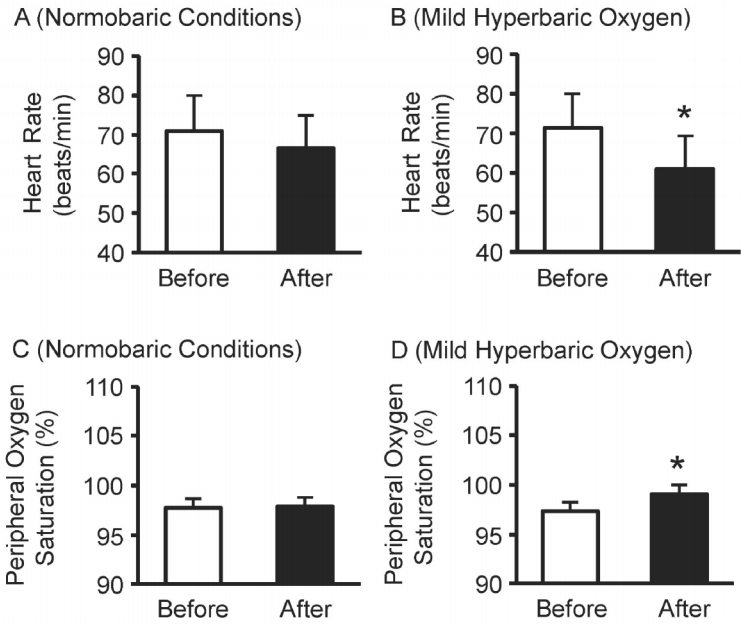

Fig. 1. Heart rate (A and B) and peripheral oxygen saturation (C and D) under normobaric (A and C) and mild hyperbaric (B and D) conditions

*Values are means and standard deviations obtained from 14 participants; *P < 0.05*

3.3 Blood Flow

Fig. 2 shows the blood flow of a participant under normobaric (top trace) and mild hyperbaric (bottom trace) conditions for 50 min. There was no change in blood flow after exposure to normobaric conditions (Fig. 3A). In contrast, the blood flow effectively doubled (from 2.7±0.5 mL/min/100 g tissue to 5.3±0.8 mL/min/100 g tissue) after exposure to mild hyperbaric oxygen (Fig. 3B).

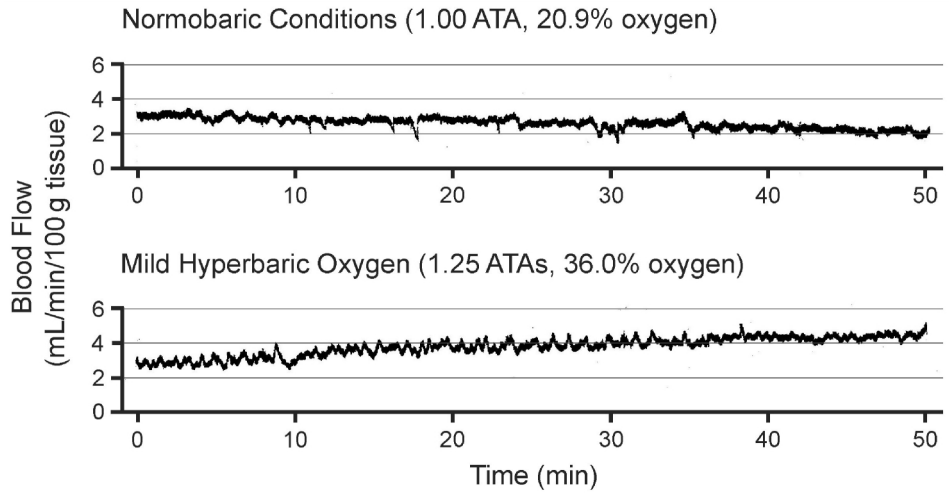

Fig. 2. Blood flow of a representative participant under normobaric (top) and mild hyperbaric (bottom) conditions for 50 min

3.4 Resting Energy Expenditure

There was no change in resting energy expenditure after exposure to normobaric conditions (Fig. 3C). In contrast, the resting energy expenditure increased by 10.2% (from 1181±125 kcal to 1296±121 kcal) after exposure to mild hyperbaric oxygen (Fig. 3D).

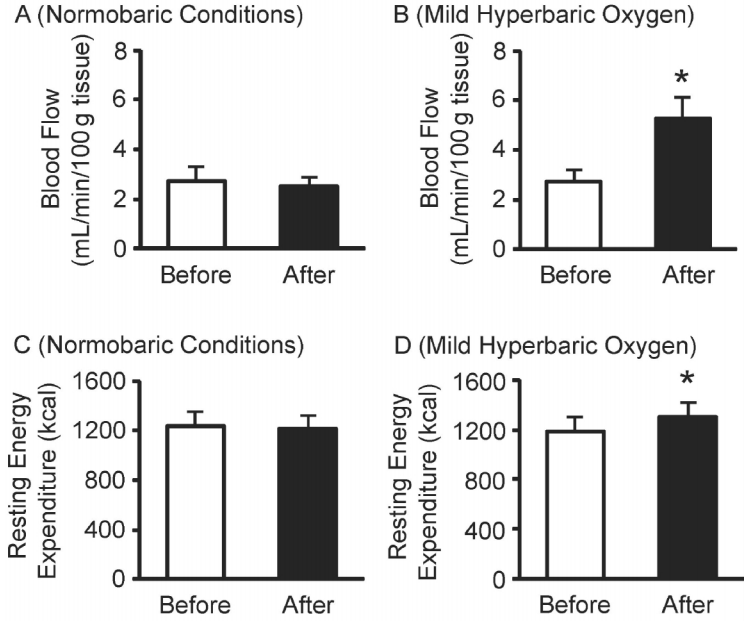

Fig. 3. Blood flow (A and B) and resting energy expenditure (C and D) under normobaric (A and C) and mild hyperbaric (B and D) conditions

*Values are means and standard deviations obtained from 14 participants; *P < 0.05*

3.5 dROMs and BAP

There was no change in the level of dROMs or BAP after either exposure to normobaric conditions or mild hyperbaric oxygen (Fig. 4).

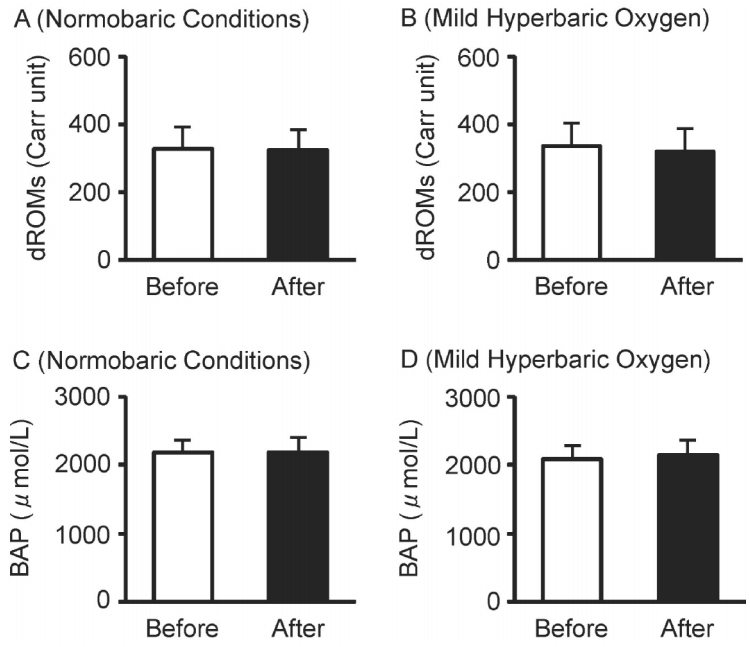

Fig. 4. Derived-Reactive oxygen metabolites (dROMs; A and B) and biological antioxidant potential (BAP; C and D) under normobaric (A and C) and mild hyperbaric (B and D) conditions

Values are means and standard deviations obtained from 14 participants

4. DISCUSSION

Hyperbaric oxygen therapy leads to vasoconstriction and hyperoxygenation, making it an effective treatment option for patients with various clinical disorders such as severe carbon monoxide poisoning, decompression sickness, and arterial gas embolism, and as adjunctive therapy for the prevention and treatment of osteoradionecrosis, clostridial myonecrosis, and compromised skin grafts and flaps [1,2]. During hyperbaric oxygen therapy, patients are generally exposed to 2–3 ATAs with 100% oxygen. However, a previous study [25] reported that exposure to hyperbaric conditions (2.5 ATAs with 100% oxygen for 2–2.5 h, 3 times per week, up to 100 times) induced cataracts in 17- to 18-month-old guinea pigs. Similarly, myopia and cataracts developed in human lenses after exposure to prolonged hyperbaric conditions at 2–2.5 ATAs with 100% oxygen for 1.5 h, once per day, from 150 to 850 times [26], although rarely after only 48 times [27]. Therefore, exposure to hyperbaric conditions at 2–3 ATAs with 100% oxygen has the potential to induce and accelerate myopia and cataracts. In addition, standard hyperbaric oxygen therapy is thought to cause excessive production of reactive oxygen species in several tissues and organs [8,28], suggesting that oxidative stress induced by hyperbaric oxygen therapy may accelerate tissue damage. Oxidative stress occurs when the production of oxidants exceeds the capacity to neutralize them, and oxidative stress levels resulting from exposure to hyperbaric conditions depend

not only on pressure but also on the duration of the exposure; a pressure of exposure of 2.5–3 ATAs and a duration of exposure of 90–120 min result in a pronounced increase in the level of oxidative stress [29,30].

We determined that exposure to mild hyperbaric conditions at 1.25 ATAs with 36.0% oxygen is sufficient to obtain effective responses in oxidative capacity in cells and tissues [9,10]. However, there are no data available concerning the response of oxidative stress and/or the levels of reactive oxygen species in patients exposed to mild hyperbaric oxygen. Therefore, in this study, we examined dROMs as an index to ascertain the level of oxidative stress in healthy humans exposed to mild hyperbaric oxygen. We found that there were no changes in the dROMs after exposure to mild hyperbaric oxygen (Fig. 4B). Interestingly, our previous study [19] showed that exposure to mild hyperbaric oxygen was effective at reducing levels of oxidative stress and C-reactive protein, which were pronounced as the result of type II collagen-induced arthritis in rats. Similarly, high blood pressure and enhanced levels of oxidative stress in spontaneously hypertensive rats were reduced by exposure to mild hyperbaric oxygen [18]. Increased sympathetic activation in hypertensive rats is mediated by the overproduction of toxic reactive oxygen species [31]. Therefore, a reduction in oxidative stress may underlie the decrease in high blood pressure observed in hypertensive rats exposed to mild hyperbaric oxygen. The results of this study using human participants, combined with the previous findings using experimental animals [18, 19], lead to the conclusion that exposure to mild hyperbaric oxygen does not affect levels of oxidative stress.

Our previous studies [9,10,13–20] using experimental animals demonstrate that exposure to mild hyperbaric oxygen increases and improves metabolism in cells and tissues. In one study [10], developing rats exposed to mild hyperbaric oxygen exhibited greater voluntary running activities compared with animals maintained under normobaric conditions, and the oxidative enzyme activities in fibers of the soleus and plantaris muscles and in motoneurons of the spinal cord that innervate these skeletal muscles increased after exposure to mild hyperbaric oxygen. Another study [20] found that an age-related decrease in oxidative capacity of skeletal muscles (e.g., a decreased percentage of high-oxidative fibers and reduced oxidative enzyme activity in the tibialis anterior muscles) of mice was reversed by exposure to mild hyperbaric oxygen. Interestingly, the skeletal muscles of Goto–Kakizaki rats with nonobese type 2 diabetes have a low oxidative capacity compared with those of nondiabetic rats [32]. In addition, the skeletal muscles of obese Long–Evans Otsuka Tokushima fatty and Zucker diabetic fatty rats, which are both type 2 diabetes animal models, contain a lower percentage of high-oxidative fibers [33,34]. These findings suggest that the low oxidative capacity of skeletal muscles in rats with type 2 diabetes may be associated with insulin resistance and impaired glucose metabolism. The growth-associated increase in blood glucose levels in rats with type 2 diabetes was attenuated by exposure to mild hyperbaric oxygen [13–15]. Furthermore, exposure to mild hyperbaric oxygen reduced high blood glucose levels and improved oxidative capacity in the skeletal muscles of adult rats with type 2 diabetes, and these effects were maintained under subsequent normobaric conditions [16].

In this study, the amount of dissolved oxygen was estimated to have increased by 2.76 times (0.864 mL/dL after exposure to mild hyperbaric oxygen/0.313 mL/dL under normobaric conditions) after exposure to mild hyperbaric oxygen. The amount of dissolved oxygen was estimated as follows: under normobaric conditions (1.00 ATA and 20.9% oxygen), the partial pressure of oxygen in the alveolus = (760 − 47) × 0.209 – 40 / 0.8 + (40 × 0.209) × (1 – 0.8) / 0.8 = 101.11 mmHg; therefore, the amount of dissolved oxygen under normobaric conditions is 0.0031 mL/dL/mmHg × 101.11 mmHg = 0.313 mL/dL. In contrast, under mild hyperbaric

conditions, the partial pressure of oxygen in the alveolus = (950 – 47) × 0.360 – 40 / 0.8 + (40 × 0.360) × (1 – 0.8) / 0.8 = 278.68 mmHg; therefore, the amount of dissolved oxygen under mild hyperbaric conditions is 0.0031 mL/dL/mmHg × 278.68 mmHg = 0.864 mL/dL, where 1.00 ATA = 760 mmHg, 1.25 ATAs = 950 mmHg, water vapor pressure = 47 mmHg, the concentration of carbon dioxide in the alveolus = 40 mmHg, and the respiratory exchange ratio = 0.8.

This study showed that the blood flow in human participants effectively doubled after exposure to mild hyperbaric oxygen (Fig. 3B). It is widely known that endurance exercises cause a steady increase in blood flow. However, the increase in blood flow following an endurance exercise is induced mostly in active skeletal muscles but not in internal organs. In addition, because of the increased atmospheric pressure, exposure to mild hyperbaric oxygen has an advantage in that it can increase the amount of dissolved oxygen in plasma, which does not occur with endurance exercises. This study also found that the resting energy expenditure in participants increased by 10.2% after exposure to mild hyperbaric oxygen (Fig. 3D).

5. PERSPECTIVES AND OPERATIVE APPLICATIONS

We previously observed in animal experiments that lifestyle-related diseases (e.g., type 2 diabetes [13–16], diabetes-induced cataract [17], and hypertension [18]) were inhibited and/or improved by exposure to mild hyperbaric oxygen. Based on the previous findings from animal models [13–18] and our observations of increased blood flow and resting energy expenditure in participants in this study, we suggest that exposure to mild hyperbaric oxygen may be an effective therapy for patients with type 2 diabetes and/or hypertension. In the future, we intend to study the effects of exposure to mild hyperbaric oxygen on disrupted nervous functions (e.g., autonomic ataxia, emotional instability, and/or dementia).

6. CONCLUSION

We conclude that exposure to mild hyperbaric oxygen increases blood flow and metabolism without increasing levels of oxidative stress.

CONSENT

Participation in the study was voluntary. Each participant received an explanation as to the aims of the study and methods of data collection and signed an informed consent form.

ETHICAL APPROVAL

All authors declare that all experiments have been examined and approved by the Institutional Experiment Committee of Kyoto University (Kyoto, Japan) and have been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

COMPETING INTERESTS

This paper has not been presented previously in any form. No conflicts of interest have been reported by the authors or by any individuals in control of the content of this paper. There were no funding or financial benefits to the authors.

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