Reducing Body Fat with Altitude Hypoxia Training in Swimmers: Role of Blood Perfusion to Skeletal Muscles

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Abstract

Swimmers tend to have greater body fat than athletes from other sports. The purpose of the study was to examine changes in body composition after altitude hypoxia exposure and the role of blood distribution to the skeletal muscle in swimmers. With a constant training volume of 12.3 km/day, young male swimmers (N = 10, 14.8 ± 0.5 years) moved from sea-level to a higher altitude of 2,300 meters. Body composition was measured before and after translocation to altitude using dual-energy X-ray absorptiometry (DXA) along with 8 control male subjects who resided at sea level for the same period of time. To determine the effects of hypoxia on muscle blood perfusion, total hemoglobin concentration (THC) was traced by near-infrared spectroscopy (NIRS) in the triceps and quadriceps muscles under glucose-ingested and insulin-secreted conditions during hypoxia exposure (16% O_2) after training. While no change in body composition was found in the control group, subjects who trained at altitude had unequivocally decreased fat mass (-1.7 ± 0.3 kg, -11.4%) with increased lean mass (+0.8 ± 0.2 kg, +1.5%). Arterial oxygen saturation significantly decreased with increased plasma lactate during hypoxia recovery mimicking 2,300 meters at altitude (~93% versus ~97%). Intriguingly, hypoxia resulted in elevated muscle THC, and sympathetic nervous activities occurred in parallel with greater-percent oxygen saturation in both muscle groups. In conclusion, the present study provides evidence that increased blood distribution to the skeletal muscle under postprandial condition may contribute to the reciprocally increased muscle mass and decreased body mass after a 3-week altitude exposure in swimmers.

Key Words: nutrient partitioning effect, adolescence, NIRS, oxygenation, body fat

Introduction

Competitive swimmers normally carry out a training volume of several kilometers per day in training, expending massive amounts of calories. However, their average body fat levels are not lower than other

types of athlete and active non-athletes (7, 12). Swimmers consuming excessive calories, thereby causing a positive energy balance, which leads to greater fat accumulation has been excluded as some well-controlled studies comparing that compare body composition across sports disciplines have shown (12, 18). Still, it is essential to understand the regulatory mechanism of body composition so as to develop effective training programs for swimmers to maintain an optimal body fat composition as a buoyancy aid without any compromise to muscle mass.

In animal studies, the effects of prolonged systemic hypoxia exposure on reducing body fat mass (6) and increasing muscle mass (10) has been established. Since ambient oxygen for fatty acid oxidation is less available under hypoxia, fat oxidation would not be the best explanation for the body fat-reducing effect of prolonged hypoxia exposure. Here, we considered that greater distribution of circulatory insulin to the skeletal muscle could directly alter the deposition of fuel storage (3), and thus can influence body composition. Insulin is the main anabolic hormone which stimulates fuel storage and syntheses of glycogen, triglyceride and protein in the skeletal muscle and adipose tissues (29). Therefore, preferential distribution in circulatory insulin perfusion among tissues would be expected to alter body composition. This is evidenced by the fact that withdrawal of insulin in animals for only 7 days can lead to marked reductions in fat mass and fat-cell diameter at all depots (13). Such conditions of shifting insulin delivery can presumably be maneuvered by increasing blood distribution towards the skeletal muscle. The role of skeletal muscles in the regulation of fat mass is demonstrated by interventions that stimulate muscle growth, which causes substantial reductions in body fat mass (1).

Due to limitations in assessments of body composition, reliable data are sparse in proving the beneficial effects of altitude hypoxia on body composition in humans. We hypothesize that: [1] a 3-week altitude exposure at 2,300 meters can increase lean mass and decrease fat mass in trained swimmers; [2] hypoxia can enhance blood distribution towards exercised skeletal muscles under postprandial insulin-stimulated conditions. To test the first hypothesis, 10 adolescent male swimmers moved from sea level to higher altitude for a 3-week stay after baseline measurement of body composition, using dual-energy X-ray absorptiometry (DXA) as an instrument for body composition measurements. To test the second hypothesis, blood hemoglobin distribution, mirroring blood distribution to skeletal muscles was traced, using near infrared spectroscopy (NIRS)under glucose-ingested hyperinsulinemic conditions during recovery from training under a simulated altitude hypoxia condition.

Materials and Methods

Subjects and Design

The present study contained two separate experiments. For the first experiment, the effects of a 3-week altitude training on body composition were determined by DXA in 10 male swimmers (age: 14.9 ± 0.4 years, BMI: 20.8 ± 0.4 kg/m^2) who regularly accomplished a total training distance of 12.3 km per day with the same volitional training effort at sea level; all subjects were motivated to complete their daily swimming volume in the shortest possible time. After baseline measurements, all subjects moved from sea level (Singapore) to altitude (2,300 m, Kunming, Yunnan, PRC) for 3 weeks. An additional 8 male control subjects (age: 13.0 ± 1.6 years, BMI: 20.5 ± 1.2 kg/m^2) residing at sea level were recruited to monitor possible time-to-time variations of DXA measurements on body composition. For the second experiment, effects of hypoxia (at 16% oxygen) on blood distribution to the skeletal muscle were assessed under glucose-ingested condition (*i.e.* insulin-stimulated condition) after training at sea level. Skeletal muscle blood distributions were measured using near infrared spectroscopy (NIRS) to detect changes in hemoglobin concentrations under hypoxic (16% oxygen) and normoxic conditions for 90 min after oral glucose ingestion. The institutional review board approved the study, and written informed parental consent and subject assent were obtained prior to any data collection.

Body Composition

Muscle and fat mass were determined by DXA before and after the 3-week altitude exposure (within 3 days upon returning to sea level). DXA is an accepted method used by the European Working Group on Sarcopenia in Older People for research and for clinical use to distinguish fat and muscle tissues (9). The whole-body DXA unit (Hologic QDR Series, Discovery W, Hologic Inc., Bedford, MA, USA) was used. It was equipped with the patented Hologic continuous calibration system and was operated by a trained radiation technician who performed all the scans. The DXA machine was calibrated every morning during the test days using a standard with a reproducibility coefficient of r = 0.90. All data were generated from the Hologic computer software (Version 9.8). DXA is based on the principle of differential attenuation of two different X-ray energies (high: 140 kVp; low 100 kVp). Participants were required to remove all metallic elements from the body and lay supine on the table, arms by the side with the palms pronated. The hips were internally rotated

Table 1. Changes in body composition after 3-week altitude exposure

	Control (N = 8)			Altitude (N = 10)		
	Pre	*Post*	*Change*	*Pre*	*Post*	*Change*
Height (m)	1.60 ± 0.22	–	–	1.72 ± 0.09	–	–
Body mass (kg)	51.9 ± 12.8	52.1 ± 13.1	0.3%	61.4 ± 6.4	61.5 ± 6.7	0.1%
BMI (kg/m^2)	20.5 ± 3.4	20.5 ± 3.4	0.1%	20.8 ± 1.3	20.8 ± 1.3	0.1%
Lean mass (kg)	39.5 ± 14.6	39.4 ± 15.1	-0.2%	48.9 ± 8.0	49.7 ± 8.0	1.5%*
Fat mass (kg)	10.7 ± 5.0	10.8 ± 4.7	1.9%	16.6 ± 5.4	14.9 ± 5.7	-11.4%*

In the table, means ± SD is shown. Asterisk (*) denotes significant difference against Pre, $P < 0.05$. Pre: before altitude translocation; Post: after 3-week altitude residence.

until both the great toes touched. The ankles were secured with velcro straps. The participants were advised to lie as still as possible during the scanning process which took about 7 min. Each participant received 0.02 mrem of radiation dose, equivalent to about 5% of the dose of a usual chest X-ray.

NIRS Measurement

NIRS is now a well-accepted method to estimate regional blood flow and oxygenation in tissues (14, 22). In the study, changes in skeletal muscle blood perfusion under hypoxia and normoxia were monitored by tissue hemoglobin level in triceps and quadriceps muscles using NIRS. NIRS measurements were performed using a frequency-domain tissue spectrometer (ISS, Champaign, IL, USA), which includes two laser diodes for measurement with wavelengths of 690 and 830 nm. The optodes were placed over the targeted muscles with the sampling rate of 1 per second. NIRS enables noninvasive continuous measurement of changes in the concentration of oxygenated hemoglobin (HbO$_2$) and deoxygenated hemoglobin (deoxyHb). Changes in the tissue concentration of total hemoglobin (THC) were calculated from the sum of changes in HbO$_2$ and deoxyHb, which is used to indicate blood distribution for the present study. Tissue oxygen saturation refers to the ratio of oxygenated hemoglobin to total hemoglobin in the microcirculation of a volume of illuminated tissue. NIRS parameters were measured in micromolar units.

Glucose and Insulin

Plasma glucose and insulin were assessedduring NIRS measurement following 75-gram glucose ingestion with 500 ml of tap water. Blood samples were collected from the fingertip at 0 (fasting value), 30, 60 and 90 min. A glucose analyzer (Lifescan, Milpitas, CA, USA) was utilized for glucose concentration determination. Insulin was determined on the ELISA analyzer (Tecan Genios, Salzburg, Austria) (5) with

the use of commercially available ELISA kits (Diagnostic Systems Laboratories, Inc. Webster, TX, USA), according to the manufacture's instruction.

Statistical Analysis

SPSS was used for all statistical analyses. Since most of baseline mean values are statistically not different, student t test was used to distinguish the mean between two trials for all measures. A level of $P < 0.05$ was set for significance for all tests, and all values are expressed as means ± standard deviation.

Results

The body composition data of swimmers measured following the 3-week altitude training are shown in Table 1. For the first experiment, the baseline age and body composition between the swimmers and control subjects were not significantly different ($P > 0.05$). The body mass and BMI of the swimmer and control groups were not significantly altered before and after the 3-week altitude exposure. However, the subjects' fat mass was significantly decreased and lean mass was significantly increased to different extents for all ten swimmers. The Control subjects who resided at sea level for the same period of time had no alterations on both measures using the same DXA equipment, which demonstrated that the above body composition changes after the altitude training were not due to time variation of the measurements.

For the second experiment, arterial oxygen saturation level after training was significantly lower in the hypoxia trial than those in the control trial (Fig. 1). The hypoxia effect was well-reflected by significantly greater blood lactate concentration above the control level (Fig. 2). Under the fed-condition, glucose and insulin levels in circulation were both elevated under the hypoxia condition after training (Fig. 3). To determine whether altitude hypoxia was the factor that caused the alterations in body composition *via* glucose and insulin redistribution, the
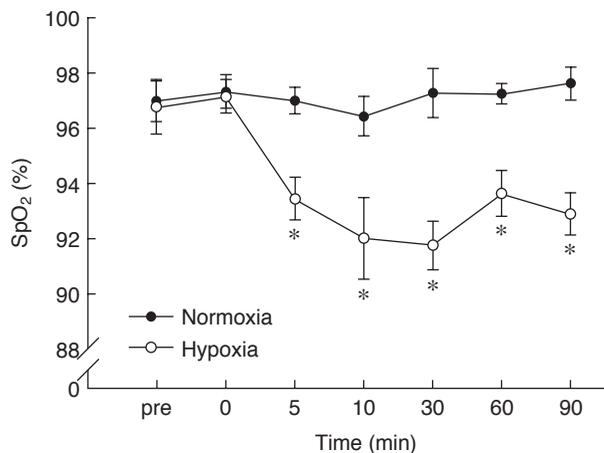
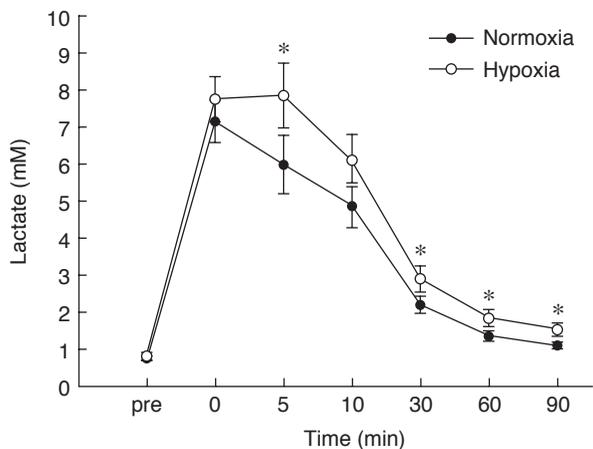Fig. 1. Arterial oxygen saturation during hypoxia recovery. Asterisk (*) denotes significant difference, $P < 0.05$.

Fig. 2. Lactate concentration during hypoxia recovery. Asterisk (*) denotes significant difference, $P < 0.05$.
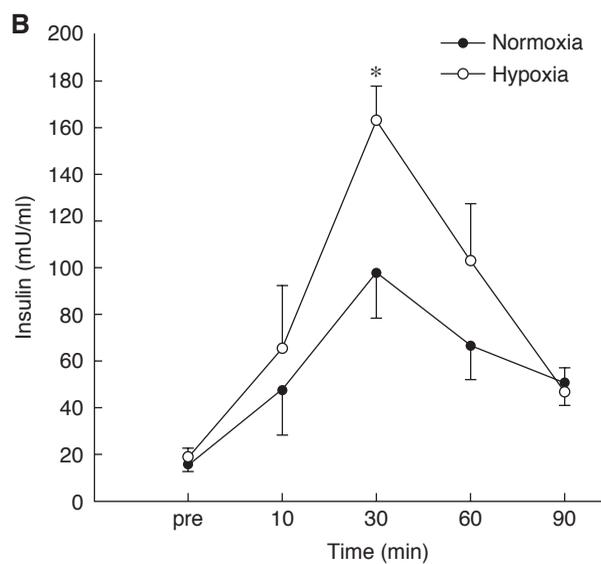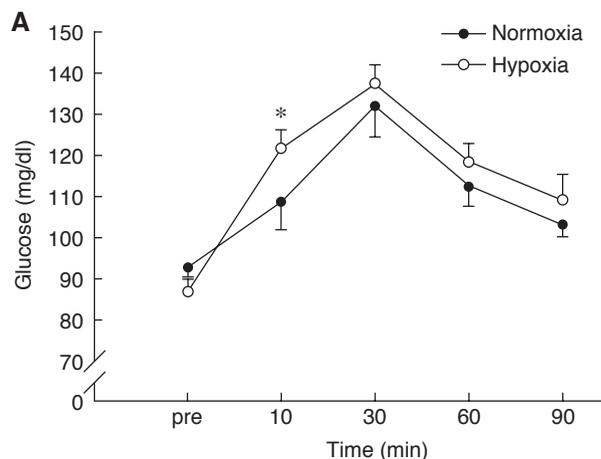

Fig. 3. Glucose and insulin changes after oral glucose consumption during hypoxia recovery. Hypoxia increased glucose (A) and insulin (B) curves above control levels. Asterisk (*) denotes significant difference, $P < 0.05$.

hemoglobin changes in both the triceps and quadriceps muscles under hypoxia were measured after glucose ingestion (Figs. 4 and 5). We found that the hypoxia exposure significantly increased total hemoglobin (A) in the skeletal muscle above the normoxic level, concurrently with greater saturation of tissue oxygen (B) on both exercised muscles than in the normal air exposure trial. This increase was paralleled with oxy-hemoglobin elevation (C). Furthermore, hypoxia did not cause changes in deoxy-hemoglobin levels (D).

Since autonomic nervous activity is directly associated with peripheral vascular control, we also determined vagal and sympathetic powers by HRV measurement under hypoxia recovery conditions (Fig. 6). Hypoxia did not affect vagal activity (high frequency power, HP) (Fig. 6A). In contrast, a significantly increased sympathetic power (low frequency to high frequency ratio) was observed under hypoxia exposure (Fig. 6B).

Discussion

The present study provides evidence in human subjects that altitude exposure can rapidly decrease fat mass and increase lean mass in youth swimmers, who maintained the same training volume at altitude as at sea level. This fat-reducing effect of altitude translocation occurred without noticeable changes in body mass. Since the overall dietary intake was not apparently changed, energy balance per se cannot be the reason to explain this significant change. The evidence of body composition change is compelling, as all ten swimmers demonstrated reciprocal decreases in fat mass and increases in lean mass after the 3-week altitude exposure. The possibility that such a change was due to an artifact caused by time-to-time variations or regression to the mean (8) of DXA measurement
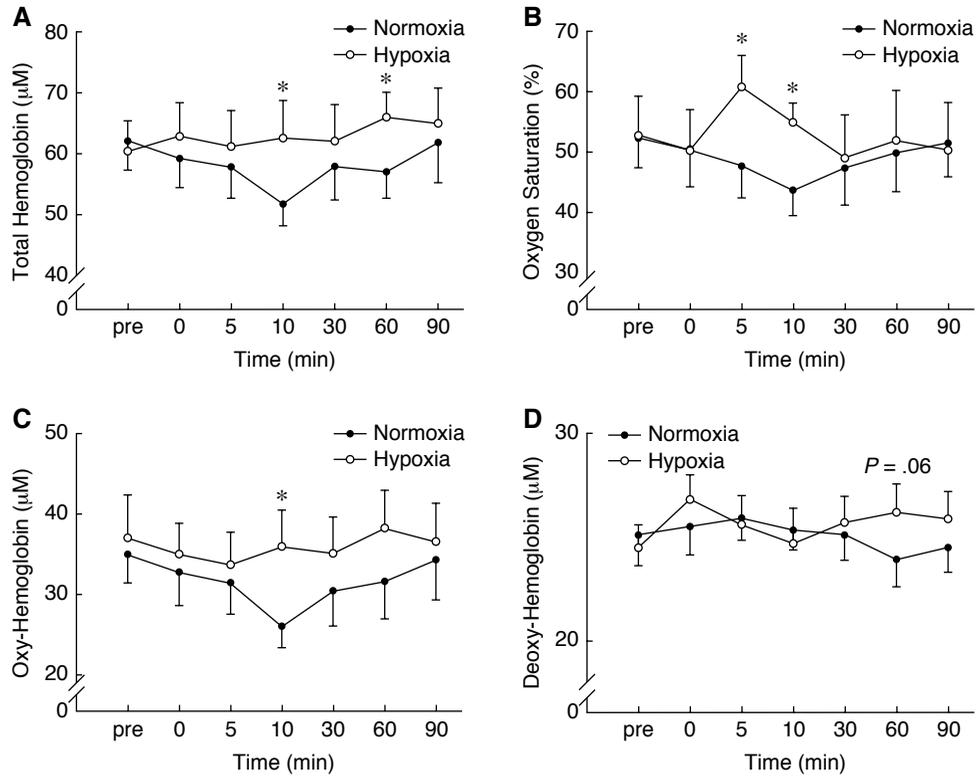
Fig. 4. Hemodynamic changes in triceps muscle during hypoxia recovery. Hypoxia significantly increases total hemoglobin (A) in the muscle above control level concurrent with increase in tissue oxygen saturation (B). This increase was paralleled with oxy-hemoglobin elevation (C). Hypoxia did not cause difference in deoxy-hemoglobin level (D).
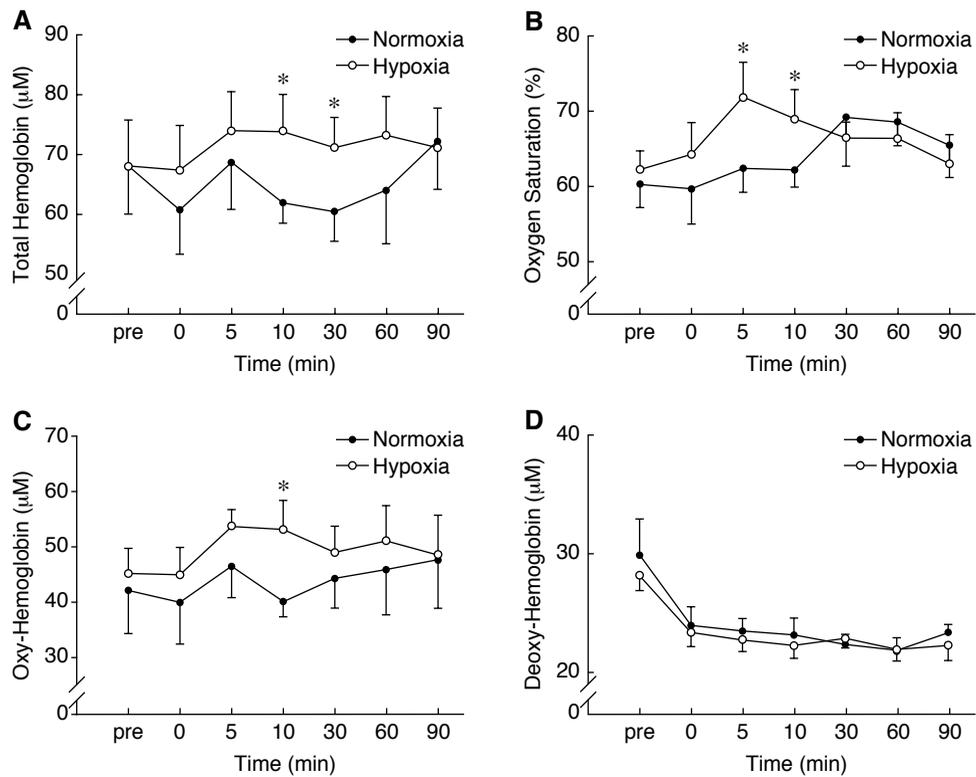
Fig. 5. Hemodynamic changes in quadriceps muscle during hypoxia recovery. Hypoxia significantly increases total hemoglobin (A) in the muscle above control level concurrent with increase in tissue oxygen saturation (B). This increase was paralleled with oxy-hemoglobin elevation (C). Hypoxia did not cause difference in deoxy-hemoglobin level (D).
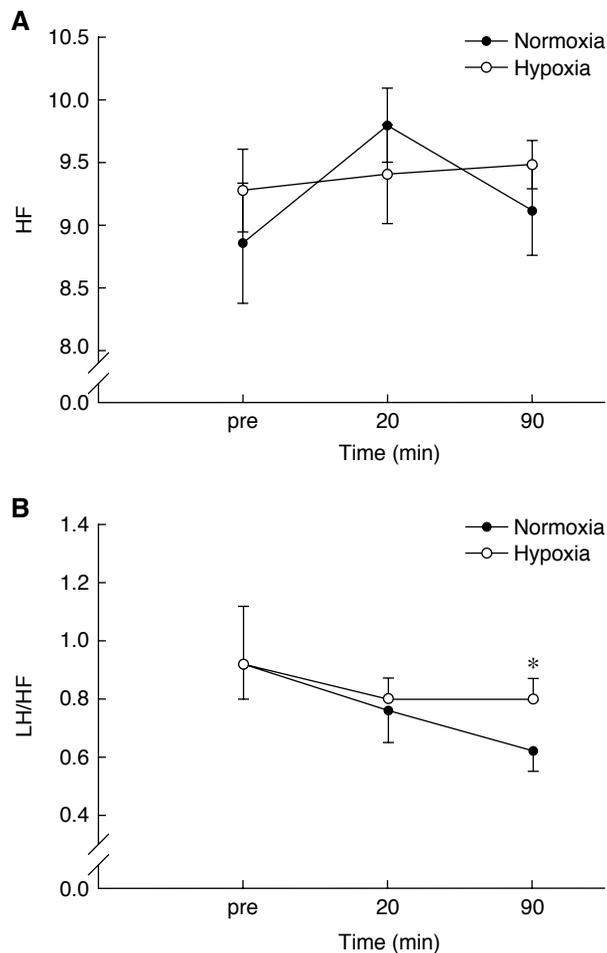
A

B

Fig. 6. Changes in autonomic nervous activity during hypoxia recovery. Hypoxia did not affect vagal activity (high frequency power, HP) (A) but significantly elevated sympathetic activity (low frequency to high frequency ratio) (B).

of muscle tissues in human body, this condition may explain, to some extent, increased muscle mass in long-term altitude exposure.

We hypothesized that hypoxia was the major contributor for the favorable change in body composition during altitude translocation, based on well-established evidence garnered from animal studies that a prolonged systemic hypoxia exposure decreased body fat (6) and increased muscle mass (10). Oxygen is the final acceptor of electron transport in fat oxidation, yet the altitude environment has relatively lower oxygen availability than at sea level for fat oxidation. Therefore, it is less likely that the observed fat-reducing effects of altitude training were mediated by increased fat metabolism at altitude.

In animals, the mechanism of increasing fuel acquisition of the skeletal muscle can lead to significant reductions in body fat mass (1, 35). In this regard, for the second part of the study, we hypothesized that the fat-reducing effect was related to increased distribution of insulin and energy substrates towards skeletal muscles in the swimmers. Our data, which showed hemoglobin increases in the skeletal muscles, fit well with the current finding of a reciprocal increase in lean mass and a decrease in fat mass for all swimmers after 3 weeks of altitude training. Increasing insulin distribution with more fuel supply into skeletal muscles can promote fuel deposition and muscle growth, which would in turn decrease fuel availability to adipose tissues. Insulin is the major anabolic hormone released after carbohydrate meals, which is able to stimulate glucose uptake (25), glycogen synthesis (4, 23), amino acid uptake (2), net protein synthesis (11), cell proliferation (24) and triglyceride synthesis (29) in both the skeletal muscle and adipose tissue (36). Therefore, competition between the skeletal muscles and adipose tissues for insulin-dependent fuel acquisition plays a key role in regulating body composition. In the present study, hemoglobin concentration in the exercised skeletal muscles was monitored under a hypoxia condition containing 16% oxygen. Using NIRS measurement, our results showed that hypoxia significantly elevated blood distribution to skeletal muscles in the absence of increased rates of arterial blood flow (BF). Although muscle BF has been shown to be elevated under systemic hypoxia (17, 28), in the present study, no significant increase in BF was observed in both muscle groups with a relatively less severe hypoxia.

The vasodilatation that occurs in the skeletal muscles during systemic hypoxia may function to enhance tissue blood distribution to offset a deficit in oxygen availability of the skeletal muscles (21, 30). Intriguingly, muscle tissue oxygen saturation, which generally mirrors the balance between oxygen demand and supply, was elevated in the subjects

are precluded, as the body composition in the control group was unchanged. The results of the study confirms those of previous animal studies (6, 10) that altitude hypoxia is an effective method to reduce body fat without compromising or negatively affecting lean mass.

During training recovery, we evaluated blood distribution under glucose-ingested hyperinsulinemic condition to mimic the postprandial state. A greater insulin levelabove normoxic condition after oral glucose challenge was observed, indicating that moderate hypoxia caused a slight insulin resistance. A similar finding has been reported elsewhere (26). This condition can likely result in an increased perfusion of both glucose and insulin preferentially into the skeletal muscle according to our NIRS data. Consistent with our observation, hypoxia has been reported to increase skeletal muscle glucose transport and vascular perfusion in rat muscles (30, 37). Considering the large amount

under hypoxia recovery. This result is in some agreement to the results of a study under a more severe hypoxia condition (lowered SaO_2 to 77 %) that muscle tissue oxygen level was not lowered by hypoxia (34).

Since vascular supply is regulated, in part, by the autonomic nervous system, the sympathetic and vagal powers by HRV were measured. Results showed that the sympathetic power was significantly elevated in the subjects under hypoxia, which might play a role in the regulation of body composition via altering blood distribution among adipose and muscle tissues. Similar results were reported under a different setting during exercise (15, 17). The role of sympathetic activity on body composition is also supported by the evidence that a prolonged use of sympathomimetic agent, such as clenbuterol, can lead to an increased muscle mass concurrently with substantial reduction in fat mass in rats (33) and humans (20). Therefore, our results suggest that the change in body composition is associated with an altered vascular control secondary to an elevation in sympathetic nervous activity. This change is expected to not only alter energy fuel distribution but also to increase insulin delivery toward the skeletal muscle. We interpret the data that fuel redistribution under hypoxia conditions can also be contributed by enhanced hepatic glucose output (19) and enhanced muscle glucose uptake (37) due, in part, to the increased sympathetic nervous stimulation.

It is surprising that the fat-reducing effect could occur within such a short period of altitude exposure (*i.e.* 3 weeks). Following exercise training, lipoprotein lipase (LPL) is released from the vascular bed, and is, therefore, redistributed in circulation (31). LPL is an enzyme which functions to hydrolyze circulating triglycerides for tissue uptake and storage. It is noteworthy that these swimmers continued the same training volume at altitude as at sea level. The increased blood distribution in the skeletal muscle suggests that LPL released from capillary bed, including the adipose tissue, was accumulated towards muscle tissues to facilitate fat deposition, while the adipose tissue becoming less likely to uptake circulating triglycerides due to competition between exercised muscle and adipose tissues for circulating factors such as oxygen, fuel and insulin. It has been shown that increasing lipoprotein lipase activity in skeletal muscles is able to decrease fat mass in training animals under the same caloric intake and exercise training load (35).

In this study, fat mass was reduced by 11.4% and muscle mass only increased 1.5% (form pre-to-post altitude exposure)The small- percent change in muscle mass is in fact large in absolute terms, given that muscle mass accounts for substantially a greater portion of the human body relative to the adipose tissue. Similar magnitude of reciprocal changes are reported in another study that measured muscle and adipose tissues directly in an exercise training setting (35). In addition, increased total energy consumption under hypoxia exposure (16) may also contribute to a greater reduction in adiposity.

We note that swimming training at moderate altitude can place swimmers under stressful conditions, which might result in impairment of cognitive function (5). More evaluation is warranted for designing a safer training-recovery protocol. Furthermore, individual variations against altitude exposure appear to vary widely and are associated with baseline DHEA-S level (27). It is current known that DHEA-S is also linked with short-term body composition change (32). It would be interesting to further determine the role of DHEA-S in body composition changes during altitude exposure.

In conclusion, swimmers are unique in having a greater fat content than most types of athletes in other sport disciplines. The current study presents an effective intervention method to reduce fat mass and increase muscle mass in youth swimmers. The reciprocal decrease in fat mass and increase in lean mass without a significant change in the body mass of the swimmers occurred after 3 weeks of training at altitude. Our evidence suggests that the preferential redistributions of insulin and nutrients towards the skeletal muscle under altitude hypoxia may be the key underlying mechanisms for the reciprocal changes in fat and lean mass of youth swimmers training at altitude.

Acknowledgments

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