

Effect of “live high, train low” strategy on stress responses in trained cyclists

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Abstract: The purpose of this study was to examine the changes in White blood cell counts (WBC) and subset counts as stress response of endurance athletes during “live high-train low” by using normobaric hypoxic room. Ten well-trained male cyclists were divided into two groups; a living in a hypoxic room and training at sea level group (LHTL group; $n=5$) and a living and training at sea level group (CONT group; $n=5$). The LHTL group stayed mainly for sleeping in a hypoxic room at a simulated altitude of 2,500m (15.4% O₂) for approximately 12 hours each day for 18 days. On the other hand, the CONT group stayed and trained in their normal living environments. Base line (Pre), and 6th, 12th and 18th day of living in the hypoxic room, WBC with subset counts (neutrophil, lymphocyte, eosinophil, basophil and monocyte counts), plasma cortisol and free fatty acid (FFA) were determined. WBC, neutrophil and basophil counts of LHTL group were significantly higher than those of CONT group on 6th day ($p<0.05$). Cortisol level of LHTL group on 12th and 18th day significantly increased from Pre ($p<0.05$), and the level on 18th day was significantly higher than that of CONT group ($p<0.05$). Our results suggested that LHTL in a normobaric hypoxic room cause the transient increase of neutrophil counts. We speculate WBC counts might be used as the handy and practical measurement of stress response by hypoxia.

Key Words: intermittent normobaric hypoxia, white blood cell counts, stress hormone

I . INTRODUCTION

Altitude training (chronic hypoxia; CH) and/or intermittent normobaric hypoxia (IH or live high train low) are considered to be effective to improve endurance capacity of athletes^(3, 7, 13, 25). However, stress reactions during altitude training may seriously cause physical conditions of athletes^(1,30). For example, many

athletes experience upper respiratory illness (URI) or gastrointestinal symptoms upon exposure to altitude. It has been suggested that this increase in the prevalence of URI and gastrointestinal symptoms may be due to an altitude-induced dysregulation of the immune system possibly caused by stress reactions. In addition, majority of athletes have been also complaining fatigue, slow recovery during alti-

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tude training or LHTL. Therefore, objective monitoring that enables stress and management will be of great help in supporting athletes in altitude training or IH.

Stress reactions during hypoxia often involve stress reactions characterized as increasing levels of blood cortisol, epinephrine, and norepinephrine^(2,5,6), as well as in altitude training (30). Stress hormones also modify the number of circulating white blood cell counts (WBC) and leukocyte subsets⁽²²⁾. Therefore, WBC and subsets counts may serve as a handy and practical measurement of stress responses during altitude training or hypoxia. However, there is no report in which stress response of athletes to LHTL was checked by WBC and subsets counts. We examined the changes in WBC and subset counts in endurance athletes during live high-train low in a normobaric hypoxic room.

II. METHOD

1. Subjects

The subjects of this study were 10 well-trained male cyclists. All of the subjects were elite cyclists who represent Japan in national and/ or international competitions or national-level university cyclists. The procedure of the study was thoroughly explained to each subject and their informed consent was obtained prior to the start of the study. This study was approved by the Juntendo University's Human Ethics Committee and complies with the Helsinki Declaration and we completed this experiment in 2005.

2. Study design

The subjects were divided into two groups, a living in a hypoxic room and training at sea level group (LHTL group; $n=5$, 25 ± 5 yr, height: 171 ± 4 cm, weight: 63.0 ± 2.2 kg) and a living and training at sea level group (CONT

group; $n=5$, age: 20 ± 1 yr, height: 171 ± 3 cm, weight: 68.8 ± 6.1 kg). During experiment, both groups performed normal volume of training. The LHTL group stayed in a hypoxic room (YHC-415, Yoshida Kankyo System, Japan) at a simulated altitude of 2,500m (15.4% O₂) for approximately 12 hours each day mainly for sleeping for 9 days and after one night at normoxia for fatigue reduction, for another 9 days. Due to the fact that subjects in the LHTL group would become acclimatized to the hypoxic exposure, and many results of our preliminary studies have shown that long-term hypoxic exposure causes fatigue in athletes, taking the practical aspect into consideration, the subjects returned to normal atmospheric conditions in 1 day on Pre, 9th and 18th day (the day before the post-values are measured) of living in the hypoxic room. The CONT group stayed and trained in their normal living environment. Oxygen concentration in the hypoxic house was recorded and monitored using by a universal recorder, TEU-10 (Espec, Japan).

3. Measurements

Before experiment (Pre; 0 day), and on 6th, 12th and 18th day of living in the hypoxic room, white blood cell counts (WBC) and subset counts (neutrophil, lymphocyte, eosinophil, basophil and monocyte counts) and plasma cortisol, free fatty acid (FFA), GOT and GPT were evaluated.

Blood samples (11 mL) of analysis were collected from the antecubital vein by a physician or a nurse at 7:00 AM on each measurement day. Of the 11 mL, 2 mL was immediately transferred to a heparinized tube subjected analyzed for WBC and subset counts; 9 mL was centrifuged at 3000rpm for 10 min and the supernatant was kept frozen at -80 °C until measurement of Cortisol, FFA, GOT and GPT at established methods. Red

blood cell counts (RBC) and Hemoglobin concentration (Hb) were collected for dehydration using the hematocrit value. During the experimental periods, resting heart rate (HR) of LHTL group was measured every morning, within 30 min after arousal.

4. Statistical analysis

All data were expressed as the mean \pm SD. Changes in the parameters before, during and after staying in the hypoxic room except resting HR were examined for significance using repeated two-way ANOVA. If a significant F value was obtained, significance difference between the different days was tested using the post-hoc test (Fisher's PLSD). The change in resting HR was analyzed by repeated one-way ANOVA followed by Dunnett Post-hoc analysis. In all cases, $p < 0.05$ was determined statistically significant.

III. RESULTS

Figure 1 shows the changes in WBC and subset counts during the experimental period. WBC, neutrophil and basophil counts of LHTL group were significantly higher than those of CONT group on the 6th day ($p < 0.05$). Basophil count of LHTL group also significantly increased from Pre. WBC and neutrophil counts tended to increase from Pre ($p <$ and $p <$, respectively). On the other hand, CONT group had higher monocyte counts as compared to LHTL group on the 6th day. Monocyte counts of both group gradually increased from Pre to 12th to 18th day ($p < 0.05$).

Cortisol level of LHTL group on 12th and 18th day significantly increased from Pre ($p < 0.05$) and the level on the 18th day was significantly higher than that of CONT group ($p < 0.05$) (Table 1). There were no changes in FFA level during the experimental period. Resting HR on 6th, 12th and 18th days had a

tendency to increase from Pre (Pre: 43.2 ± 3.5 beats/min, 6th day; 49.6 ± 4.3 beats/min, 12th day; 50.9 ± 7.8 beats/min, 18th day; 48.7 ± 5.2 beats/min)(Data not shown.).

IV. DISCUSSION

The primary findings in this study were that WBC, neutrophil and basophil counts as index of stress responses transiently increased at an early stage during LHTL, however, serum cortisol gradually increase during LHTL.

There were a few reports on changes in WBC counts and subset counts to altitude. One study reported that a rise in granulocyte counts in ascending to 4300m⁽²⁶⁾ and another an increase in certain lymphocyte subsets⁽¹²⁾. Unfortunately, to our knowledge, as for the effect of LHTL on WBC and subset counts, there has been no report on. However, these WBC and subset count relate to stress hormones. Okutsu et al.⁽²²⁾ reported that stress hormones change the number of circulating WBC and leukocyte subsets. Therefore, transitory increase in WBC, neutrophil and basophil counts may indicate stress responses to LHTL. We considered increasing of WBC counts may caused by neutrophil and basophil counts.

There are some possibilities on the factor of neutrophil counts increasing. That is, stress hormones as catecholamine come from the autonomous nervous system, cortisol and/or growth hormone secrete with physiological and mental stress^(8, 10, 11, 14, 20, 21, 23, 24). The sympathetic nervous system is responsible for the regulation of several physiological responses including heart rate, respiratory rate, and substrate utilization. Two primary mediators of the sympathetic nervous system are the catecholamines, epinephrine and norepinephrine. Several studies have demonstrated that the

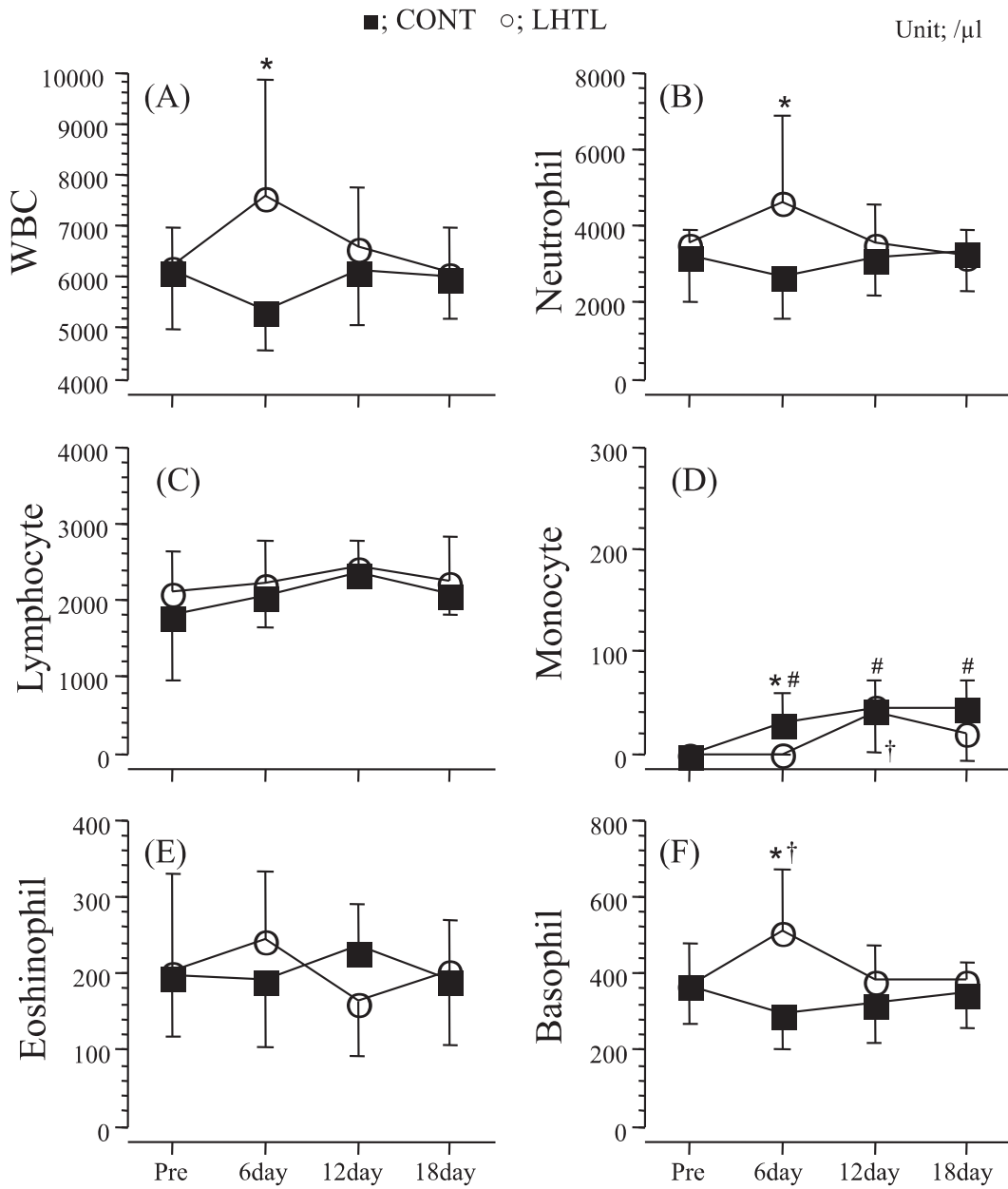


Figure 1. Changes in (A) WBC counts; (B) neutrophil counts; (C) lymphocyte counts; (D) monocyte counts; (E) eosinophil counts and (F) basophil counts for CONT and LHTL groups. Values are mean \pm SD. * Significant difference at 6th day between group ($p < 0.05$) † Significant difference from pre value in LHTL group ($p < 0.05$) # Significant difference from pre value in CONT group ($p < 0.05$).

Table 1. Change in serum cortisol and free fatty acid (FFA) for control group (CONT) and experimental group (LHTL).

		Pre	6th day	12th day	18th day
Cortisol (μ g/dl)	CONT	22.6 \pm 2.4	24.2 \pm 1.2	24.6 \pm 3.5	23.7 \pm 4.2
	LHTL	23.4 \pm 3.3	26.5 \pm 2.6	28.7 \pm 5.5 †	28.5 \pm 2.6 *†
FFA (mEq/L)	CONT	0.32 \pm 0.22	0.25 \pm 0.10	0.21 \pm 0.09	0.29 \pm 0.18
	LHTL	0.22 \pm 0.09	0.20 \pm 0.10	0.28 \pm 0.13	0.20 \pm 0.08

Values are means \pm SD. FFA; free fatty acid. * Significant difference at 6th day between groups. † Significant difference from pre value in LHTL group.

sympathetic nervous system is up-regulated at rest and during exercise in response to acute and chronic exposure to 4300m as evidenced by increased plasma and urinary levels of epinephrine and norepinephrine⁽¹⁵⁻¹⁹⁾. Hypoxia-induced increase in catecholamines, occurring within the first hours of altitude exposure but return to sea level values by the fifth day at altitude^(15, 18, 19). Although we did not measure catecholamines, FFA and resting HR as markers related to the originate in sympathetic nervous system examined in this study. Resting HR of LHTL group tended to increase and maintained high values during the experimental period, although FFA concentration did not change. Therefore, we speculate that transitory increase in neutrophil counts occurring on at 6th day in this study indicate increase of stress hormone as catecholamine.

Exposure to hypoxia and/or altitude training also caused cortisol as stress hormone⁽²⁹⁾. Wilber et al.⁽³⁰⁾ reported that serum cortisol in well-trained endurance athletes increased progressively and significantly elevated at the end of the five weeks altitude training at moderate altitude (1885m) and speculated that the significant increase in serum cortisol at the end of altitude training camp reflected the additive effect of hypobaric hypoxia for five weeks in combination with a progressively increased training load. Also, Uchakin et al.⁽²⁸⁾ investigate that effect of staying at 2500m and training at 1250m (LHTL) for four weeks on trained runners and reported a post LHTL serum cortisol concentration was significantly higher than a pre value. Our result of serum cortisol responses during LHTL was consistent with these studies. Although mechanisms of increasing serum cortisol at altitude training and/or LHTL was not clear, it seem that training stress may have affected the cortisol response to a greater degree than hypoxic stress. On the other hand, Hypoxia is known

to modify cortisol secretion, although its effect depends on the experimental conditions. For example, resting plasma cortisol concentrations have been reported to increase^(9, 20) or unchange⁽²⁷⁾ in human beings exposed to acute hypoxia. Also, Coste, et al.⁽⁴⁾ reported that prolonged mild hypoxia on circadian cortisol rhythm in human beings. Therefore, our results in cortisol responses may effect not only training but also circadian rhythm.

Furthermore, Bailey et al.⁽¹⁾ (1998) evaluated two groups of elite male distance runners who trained four weeks at either 1500 to 2000m or 1640m. They found that a 50 increase in the upper respiratory and gastrointestinal tract infections during altitude training in both groups and resting plasma glutamine concentrations decreased by 19% after three weeks at altitude. Therefore, it has been suggested that increased serum levels of stress hormones such as cortisol may be induced to an altitude-induced suppression of the immune system. We recognized increased serum levels of cortisol at the 12th and 18th days during LHTL, whereas neutrophil counts transitory increased on 6th day. Thus, we could not find that the relationship between neutrophil counts and cortisol responses.

However, in contrast, some researchers reported no change of serum cortisol on altitude training. The discrepancy in these studies of serum cortisol responses at moderate altitude may be due to differences in the hypoxic stimulus and individuality of responses. Also, stress responses to hypoxia relate to not only cortisol as stress hormone but also various factors. A prospective study is necessary to further confirm this effect.

V. CONCLUSION

From these results, it was suggested that LHTL using by normobaric hypoxic room in-

duces to increases transient neutrophil counts. We speculate WBC counts is a handy and practical measure of stress response by hypoxia. And, we might easily find athletes with stronger stress reactions and possibility to advise pre-conditioning at lower altitude or sea level.

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