Growth Hormone & IGF Research 25 (2015) 121–126

Contents lists available at ScienceDirect

Growth Hormone & IGF Research

journal homepage: www.elsevier.com/locate/ghir

Hormonal and metabolic responses to repeated cycling sprints under different hypoxic conditions

Michihiro Kon [a,b,*], Kohei Nakagaki [b], Yoshiko Ebi [b], Tetsunari Nishiyama [c], Aaron P. Russell [d]

[a] School of International Liberal Studies, Chukyo University, 101-2 Yagotohonmachi, Showa-ku, Nagoya 466-8666, Japan
[b] Department of Sports Sciences, Japan Institute of Sports Sciences, 3-15-1 Nishigaoka, Kita-ku, Tokyo 115-0056, Japan
[c] Department of Physical Education, Nippon Sport Science University, 7-1-1 Fukasawa, Setagaya-ku, Tokyo 158-8508, Japan
[d] Centre for Physical Activity and Nutrition Research, School of Exercise and Nutrition Sciences, Deakin University, 221 Burwood Hwy, 3125, Australia

ARTICLE INFO

Article history:
Received 29 December 2014
Received in revised form 29 March 2015
Accepted 29 March 2015
Available online 12 April 2015

Keywords:
Growth hormone
Lipolysis
Supramaximal exercise
Systemic hypoxia

ABSTRACT

Objective: Sprint exercise and hypoxic stimulus during exercise are potent factors affecting hormonal and metabolic responses. However, the effects of different hypoxic levels on hormonal and metabolic responses during sprint exercise are not known. Here, we examined the effect of different hypoxic conditions on hormonal and metabolic responses during sprint exercise.
Design: Seven male subjects participated in three experimental trials: 1) sprint exercise under normoxia (NSE); 2) sprint exercise under moderate normobaric hypoxia (16.4% oxygen) (HSE 16.4); and 3) sprint exercise under severe normobaric hypoxia (13.6% oxygen) (HSE 13.6). The sprint exercise consisted of four 30 s all-out cycling bouts with 4-min rest between bouts. Glucose, free fatty acids (FFA), blood lactate, growth hormone (GH), epinephrine (E), norepinephrine (NE), and insulin concentrations in the HSE trials were measured before exposure to hypoxia (pre 1), 15 min after exposure to hypoxia (pre 2), and at 0, 15, 30, 60, 120, and 180 min after the exercise performed in hypoxia. The blood samples in the NSE trial were obtained in normoxia at the same time points as the HSE trials.
Results: Circulating levels of glucose, FFA, lactate, GH, E, NE, and insulin significantly increased after all three exercise trials ($P < 0.05$). The area under the curve (AUC) for GH was significantly higher in the HSE 13.6 trial than in the NSE and HSE 16.4 trials ($P < 0.05$). A maximal increase in FFA concentration was observed at 180 min after exercise and was not different between trials.
Conclusion: These findings suggest that severe hypoxia may be an important factor for the enhancement of GH response to all-out sprint exercise.

1. Introduction

Exercise is a potent stimulus for growth hormone (GH) secretion. The GH response to acute exercise is related to exercise intensity with greater GH secretion observed at higher exercise intensities [1]. Although the precise physiological role of high intensity exercise-stimulated GH secretion has not been fully elucidated, it has been associated with a post-exercise rise in lipolysis [2]. Octreotide infusion blocks exercise-induced GH secretion and inhibits post-exercise adipose tissue lipolysis [3]. These results suggest that high intensity exercise-induced GH release enhances adipose tissue lipolysis during the post-exercise recovery period.

Exercise performed in acute severe systemic hypoxia also increases GH release [4–6]. We have recently reported that moderate- [4] and low- [5] intensity resistance exercise performed during acute severe hypoxia (13% oxygen) causes a larger increase in GH concentrations than when exercise is performed in normoxia. Additionally, submaximal endurance (cycle ergometer) exercise (750 kpm/min) performed under severe hypoxic conditions (4550 m above sea level; approx. 12% oxygen) increases levels of circulating GH and free fatty acids (FFA) to a greater extent than when exercise is performed under normoxic conditions [6]. In contrast, low-intensity (30% 1RM) resistance exercise performed under moderate hypoxia (15% oxygen) or normoxia caused similar levels of GH secretion [7]. Similarly, 30 min of submaximal endurance exercise (50% of peak oxygen uptake) performed under moderate hypoxia (2000 m above sea level; approx. 16.5% oxygen) or normoxia elicited similar GH and FFA responses [8]. These results suggest that the severity

Abbreviations: NSE, sprint exercise under normoxia; HSE 16.4, sprint exercise under moderate normobaric hypoxia (16.4% oxygen); HSE 13.6, sprint exercise under severe normobaric hypoxia (13.6% oxygen); FFA, free fatty acids; GH, growth hormone; E, epinephrine; NE, norepinephrine; AUC, area under the curve; 1RM, one-repetition maximum; BMI, body mass index; SpO_2, arterial oxygen saturation.

* Corresponding author at: School of International Liberal Studies, Chukyo University, 101-2 Yagotohonmachi, Showa-ku, Nagoya 466-8666, Japan. Tel.: +81 52 835 9864; fax: +81 52 835 7197.
E-mail address: kon.michihiro@gmail.com (M. Kon).

http://dx.doi.org/10.1016/j.ghir.2015.03.002
1096-6374/

of hypoxia affects the exercise-induced GH secretion and adipose tissue lipolysis, and subsequent FFA mobilization.

All-out sprint cycling exercise is a powerful stimulus for GH release. Previous studies report a marked increase in circulating GH levels following sprint exercise [9–12]. However, as far as we know, there are no data concerning the effect of different hypoxic levels on circulating GH and FFA responses during and post-sprint exercise. Improved repeated sprint ability performance in athletes occurs following sprint exercise training under hypoxia [13]. Indeed athletes, in particular international-level athletes, utilize hypoxic training rooms to gain a greater improvement in their physical performance. A reduction in body fat can further enhance athlete physical performance. Therefore, a better understanding of the relationship between hypoxia and circulating GH and FFA responses during and post-sprint exercise may assist in improving athlete performance. Here, we examined the effects of sprint exercise performed under both moderate and severe hypoxic conditions on hormonal and metabolic responses. We hypothesized that following sprint exercise performed in severe hypoxic conditions GH and circulating FFA levels would be elevated to a greater extent when compared to the sprint exercise performed in normoxic and moderate hypoxic conditions.

2. Methods

2.1. Subjects

Seven healthy male subjects (age = 20.4 ± 0.7 yr, height = 170.4 ± 1.5 cm, body mass = 68.3 ± 1.9 kg, BMI = 23.5 ± 0.9 kg/m^2) participated in this study. The subjects were non-smokers and not taking regular medications. They belonged to a University Cycling Club and trained 6 days per week. The subjects were not exposure to an altitude of >3000 m within 1 month before each trial and had no history of severe acute mountain sickness. All subjects were informed about the purpose of the present study as well as the experimental procedures and their written informed consent was obtained in advance. The study was approved by Japan Institute of Sports Sciences Ethics Committee.

2.2. Exercise trials

This study was conducted using a single-blinded design. All subjects participated in three experimental trials at least 10 days apart: 1) sprint exercise under normoxia (NSE), 2) sprint exercise under moderate normobaric hypoxia (16.4% oxygen) (HSE 16.4), and 3) sprint exercise under severe normobaric hypoxia (13.6% oxygen) (HSE 13.6). The sprint exercise consisted of four 30 s 'all out' cycling bouts on a cycle ergometer (Power Max VII, Combi Co., Ltd., Tokyo, Japan) at a resistance equivalent to 7.5% of their body mass with 4 min recovery (rest) between bouts. Subjects were instructed to sprint as fast as possible against the ergometer's resistance and were encouraged verbally. Peak power and mean power of each bout were measured and recorded. The trials were performed between 07:30 am and 12:30 pm. All trials were performed in the strength and conditioning facilities at the Japan Institute of Sports Sciences (Tokyo, Japan). The hypoxic sprint exercises (HSE 16.4 and HSE 13.6) were conducted in a specially designed hypoxic training room within the facility. The hypoxic environment was created by filtering compressed air through a high-polymer membrane [14].

2.3. Blood sampling and analyses

The subjects came to the laboratory 2.5 h after consuming the same light meal (180 kcal) and rested for 30 min before the first blood collection. Venous blood samples in the HSE trials were obtained from each subject's forearm before exposure to hypoxia (pre 1), 15 min after the exposure to hypoxia (pre 2), and at 0 (immediately after the exercise), 15, 30, 60, 120, and 180 min after exercise in hypoxia. The blood samples in the NSE trial were obtained in normoxia at the same time points as the HSE trials. Exposure to the hypoxic conditions continued until the experimental trials ended (180 min after exercises). Plasma and serum were separated from blood cells by centrifugation (3000 rpm for 15 min) and stored at −80 °C until analysis. Plasma glucose concentration was measured using an enzymatic method with a commercial kit (LSI Medience Co., Tokyo, Japan). The intra-assay and inter-assay coefficients of variation (CVs) were 0.22% and 1.6% for glucose, respectively. The glucose assay sensitivity was 1.0 mg/dl. Serum FFA concentration was analyzed with an enzymatic-UV method using commercially available kits (LSI Medience Co., Tokyo, Japan). The sensitivity of the FFA assay, and intra-assay and inter-assay CVs was 0.04 mEq/l and 0.34% and 2.75% for FFA, respectively. Serum GH concentration was measured by chemiluminescent enzyme immunoassay using a commercial kit (Beckman Coulter, Inc., Tokyo, Japan). The intra-assay and inter-assay CVs were 3.6% and 5.1% for GH, respectively. The GH assay sensitivity was 0.002 ng/ml. Plasma epinephrine (E) and norepinephrine (NE) concentrations were measured with a high-performance liquid chromatography using a commercial kit (Tosoh Co., Tokyo, Japan). The intra-assay and inter-assay CVs were 4.08% and 0.81% for E and 0.81% and 2.27% for NE, respectively. The sensitivity of these assays was 0.02 ng/ml for E and NE. Serum insulin was determined using chemiluminescent immunoassay with a commercial kit (ABBOTT JAPAN Co., Ltd., Chiba, Japan). The sensitivity of the insulin assay, and intra-assay and inter-assay CVs was 0.5 μU/ml and 3.9% and 2.9% for insulin, respectively. Blood samples were obtained from the fingertip to measure blood lactate concentration using an automatic lactate analyzer (Lactate Pro; ARKRAY, Kyoto, Japan). Arterial oxygen saturation (SpO_2) was measured by pulse oximetry from the second finger (PULSOX-Me300; Tenjin Ltd., Osaka, Japan).

2.4. Statistical analysis

All data were analyzed by a two-way ANOVA with repeated measures. When required a Bonferroni/Dunn post hoc analysis was performed. A one-way ANOVA with repeated measures was used to analyze the area under the curve (AUC) that was calculated by a standard trapezoidal technique. If significant differences were noted, a post hoc analysis test (Bonferroni/Dunn) was performed. The percent changes from pre 1 between the groups were compared using a one-way ANOVA with repeated measures. If significant differences existed, a Bonferroni/Dunn post hoc analysis was used. Pearson's correlation coefficient was calculated to evaluate relationships between peak values of FFA and hormones. The level of statistical significance was set at $P < 0.05$. Data are expressed as mean ± SE.

3. Results

3.1. Power outputs

Table 1 shows the results for the peak and mean power outputs. There was a significant main effect of time on peak ($F = 47.65$,

Table 1
Changes in peak power and mean power.

		NSE	HSE 16.4	HSE 13.6
Peak power (watt)	Bout 1	845 ± 20	812 ± 18	790 ± 20
	Bout 2	804 ± 21	795 ± 19	783 ± 21
	Bout 3	751 ± 21 **	755 ± 16 **	739 ± 15**
	Bout 4	705 ± 21 **	705 ± 25 **	685 ± 22 **
Mean power (watt)	Bout 1	682 ± 13	670 ± 12	638 ± 11
	Bout 2	600 ± 10 **	622 ± 9 **	607 ± 11**
	Bout 3	567 ± 19 **	575 ± 10 **	554 ± 7 **
	Bout 4	534 ± 16 **	540 ± 23 **	504 ± 12 **

Values are represented as means ± SE (n = 7). NSE, nonnoxic sprint exercise; HSE 16.4, hypoxic (16.4%) sprint exercise; and HSE 13.6, hypoxic (13.6%) sprint exercise. ** $P < 0.01$ vs Bout 1.