Plasma Leptin Levels of Elite Endurance Runners after Heavy Endurance Training

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Abstract A decrease in testosterone levels and an increase in cortisol levels are observed in male athletes with the overtraining syndrome (OTS). Cortisol causes blood leptin levels to rise and testosterone has an inverse relationship with blood leptin levels. Therefore, we hypothesized that the hormonal changes as a result of OTS induce an increase in leptin. To test this hypothesis, we examined the relationship among changes in leptin, testosterone and cortisol in thirteen male collegiate distance runners (aged 20.3 ± 1.1 years) before and after an 8-day strenuous training camp. Runners ran 284.1 ± 48.2 km during the training camp. Body fat percentages and plasma glucose concentrations decreased significantly after the training. Non-ester fatty acids and total cholesterol concentrations in blood were unchanged. Serum cortisol concentrations showed a significant increase after the training camp (from 11.82 ± 2.00 μg/dl to 16.78 ± 3.99 μg/dl), and serum testosterone decreased significantly (from 408.0 ± 127.6 ng/dl to 265.2 ± 97.6 ng/dl). The ratio of testosterone to cortisol (TCR) dropped by 50% after training (from 35.62 ± 13.69 to 16.94 ± 8.47). These results suggest that the subjects reached a state of the OTS. Contrary to our hypothesis, plasma leptin was not significantly changed (from 1.34 ± 0.29 ng/ml to 1.49 ± 0.18 ng/ml). Δ Plasma leptin was not significantly correlated with Δ serum cortisol, Δ TCR or Δ fat percentage. However, Δ serum testosterone was positively correlated with Δ plasma leptin (r = 0.596, p < 0.05). Plasma leptin concentrations might modulate the secretion of testosterone in overtraining conditions. In conclusion, the change in blood leptin level is independent of the changes in cortisol, TCR and fat percentage in highly trained male athletes in the state of the OTS. J Physiol Anthropol Appl Human Sci 24 (6): 573-578, 2005 http://www.jstage.jst.go.jp/browse/jpa [DOI: 10.2114/jpa.24.573]

Keywords: distance runner, overtraining, the ratio of testosterone to cortisol (TCR), leptin

Introduction

Leptin is a peptide hormone secreted mainly from adipocytes that induces appetite suppression and an increase in energy expenditure via the hypothalamus (Campfield et al., 1995; Zhang et al., 1994). It is generally accepted that blood leptin levels are strongly related to body fat mass and body fat percentage in both animals and humans (Considine et al., 1996; Maffei et al., 1995).

The blood leptin level remains unchanged immediately after acute endurance exercise, but declines on the following day (Desorges et al., 2004b; Duclos et al., 1999; Elias et al., 2000; Essig et al., 2000; Jürimäe et al., 2005; Olive and Miller, 2001; Perusse et al., 1997; Torjman et al., 1999; Weltman et al., 2000; Zafeiridis et al., 2003). Chronic exercise (i.e., exercise training) reduces body fat mass, which may result in a decreased blood leptin concentration in non-athletes (Hickey et al., 1997; Houmard et al., 2000; Perusse et al., 1997). In highly trained athletes, however, blood leptin levels decrease without significant changes in body fat content after exercise training (Gomez-Merino et al., 2002; Jürimäe et al., 2003).

Chronic fatigue induced by excessive training is known as overtraining syndrome (OTS) (Smith, 2000). A typical sign of overtraining in male athletes is an increase in cortisol and a decrease in testosterone concentrations in the blood (Smith, 2000). Therefore, the ratio of testosterone to cortisol (TCR) is suggested to be a useful index of OTS in male athletes. It has been suggested that testosterone has an inhibitory effect on leptin secretion from adipose tissue in adult (Luukkaa et al., 1998) and adolescent male athletes (Blum et al., 1997; Weimann, 2002). Additionally, glucocorticoids are one of stimulators of leptin secretion from adipocytes in humans (Leal-Cerro et al., 2001). These findings imply the possibility that decreased testosterone and increased cortisol levels observed in OTS induce hyperleptinemia in male athletes. Thus, the purpose of this study was to examine the relationship between the change in blood leptin levels and the parameters of OTS (i.e., testosterone, cortisol, and TCR) in male collegiate elite distance runners after a strenuous training camp.
Materials and Methods

Subjects and experimental design

Thirteen elite collegiate male distance runners aged 20.5 ± 1.1 years gave written informed consent to participate in this experiment. Their best results (5,000 m) were 14'38"19 ± 31"43. Subjects had no practice for four consecutive days prior to the experiment. Before and after an 8-day severe training camp, height, body mass and skinfold thickness of six positions (triceps, subscapular, chest, abdomen, supraillium and thigh) were measured. Running distances and body mass were also recorded every day. Body mass index (BMI) was calculated as the body mass divided by height squared (kg/m²), and fat percentage was estimated from the following equation: Fat percentage = 0.21601X – 0.00029X² + 0.13341Y – 5.72888, where X = sum of six skinfolds, and Y = age in years (Jackson, 1982). Lean body mass (LBM) was determined by body mass minus fat mass that was calculated as body mass multiplied by percentage fat. The present study was performed in accordance with the guidelines for the Declaration of Helsinki–Ethical Principles for Medical Research Involving Human Subjects (World Medical Association).

Blood determination

Two blood samples were withdrawn from the antecubital vein between 9:00 a.m. and 10:00 a.m. on the first day of the camp and on the day after the end of the training camp. Each day, the subjects finished breakfast by 6:00 a.m.

The following biochemical parameters in blood were measured: serum non-ester free fatty acids (NEFA) (NEFA-SS EIKEN, Eiken Chemical, Tokyo, Japan), serum total cholesterol (T-Chol; DA4101, Daichii Pure Chemicals), plasma glucose concentrations (Merck Liquid Gli, Kanto Chemical, Tokyo, Japan), serum testosterone (Coat-A-Count Total Testosterone, Diagnostic Products Corporation, Los Angeles, CA), serum cortisol (Gamma Coat Cortisol, Incstar Corporation, Stillwater, MN), and plasma leptin (Human Leptin RIA, LINCO Research, St. Charles, MO). TCR was calculated by dividing the testosterone concentrations by those of cortisol.

Assessment of caloric intake

Daily dietary caloric intakes were recorded with photographs and written lists throughout the training camp. The amount of energy intake was evaluated by computer software (EXCEL EIYOUKUN Ver. 3.0, Kenpakusha, Tokyo, Japan).

Statistical analysis

Values are expressed as means ± S.D. Statistical significance was determined using a paired Student's t-test between the pre- and post-training camp values. Values were considered to be statistically significant at p < 0.05.

Results

Training volume and physical characteristics

Subjects ran 284.1 ± 48.2 km during 8 days of training camp in total. There were no significant changes in body mass, BMI and LBM between pre- and post-training camp (Table 1). However, a significant decrease in fat percentage was observed after the training camp.

Blood biochemical parameters

No significant changes in serum NEFA, serum total cholesterol, and hematocrit levels were found between before and after the training camp (Table 2). Significant decreases in plasma glucose concentrations were observed after the training camp when compared with pre-training values. The eight-day severe training camp increased plasma cortisol concentrations and decreased serum testosterone concentrations, resulting in a substantial reduction in TCR. Plasma leptin levels were not affected by the training camp.

With respect to the differences in biochemical parameters between the pre- and post-training camp, Δ plasma leptin was not significantly correlated with Δ serum cortisol or Δ TCR (Fig. 1). However, there was a positive correlation between Δ plasma leptin and Δ serum testosterone (r = 0.596, p < 0.05). In addition, the correlation between Δ plasma leptin and Δ fat percentage was insignificant after the training camp (Fig. 2).

Caloric intake

The subjects ate 4,372 kcal of food per day. The caloric ratio

Table 1 Physical characteristics of subjects (n=13) pre and post training camp

<table>
<thead>
<tr>
<th></th>
<th>Pre</th>
<th>Post</th>
<th>p&lt;</th>
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<tbody>
<tr>
<td>BMI (kg/m²)</td>
<td>19.01 ± 0.75</td>
<td>18.97 ± 0.66</td>
<td>0.3932</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>57.65 ± 2.96</td>
<td>57.54 ± 2.73</td>
<td>0.3870</td>
</tr>
<tr>
<td>Fat percentages (%)</td>
<td>3.60 ± 0.72</td>
<td>3.23 ± 0.62</td>
<td>0.0003</td>
</tr>
<tr>
<td>LBM (kg)</td>
<td>55.57 ± 2.74</td>
<td>55.47 ± 2.55</td>
<td>0.4778</td>
</tr>
</tbody>
</table>

Values are means ± S.D. BMI: Body mass index; LBM: Lean body mass.

Table 2 Biochemical parameters in blood: pre and post training camp

<table>
<thead>
<tr>
<th></th>
<th>Pre</th>
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<tbody>
<tr>
<td>Hematocrit (%)</td>
<td>45.92 ± 3.37</td>
<td>45.09 ± 2.97</td>
<td>0.2687</td>
</tr>
<tr>
<td>NEFA (mEq/l)</td>
<td>0.152 ± 0.112</td>
<td>0.229 ± 0.218</td>
<td>0.0719</td>
</tr>
<tr>
<td>Total cholesterol (mg/dl)</td>
<td>174.5 ± 20.1</td>
<td>179.1 ± 18.5</td>
<td>0.4460</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td>88.8 ± 12.1</td>
<td>75.5 ± 14.0</td>
<td>0.0066</td>
</tr>
<tr>
<td>Cortisol (µg/dl)</td>
<td>11.82 ± 2.00</td>
<td>16.78 ± 3.99</td>
<td>0.0009</td>
</tr>
<tr>
<td>Testosterone (ng/dl)</td>
<td>408.0 ± 127.6</td>
<td>265.2 ± 97.6</td>
<td>0.0028</td>
</tr>
<tr>
<td>TCR</td>
<td>35.62 ± 13.69</td>
<td>16.94 ± 8.47</td>
<td>0.0003</td>
</tr>
<tr>
<td>Leptin (ng/ml)</td>
<td>1.34 ± 0.29</td>
<td>1.49 ± 0.18</td>
<td>0.0518</td>
</tr>
</tbody>
</table>

Values are means ± S.D. NEFA: non-ester free fatty acids; TCR: the ratio of testosterone to cortisol.
of nutrition was 16.4% protein, 24.2% fat, and 59.4% carbohydrate. According to the dietary reference intake in The Sixth Revision of The Japanese Recommended Dietary Allowances (Ministry of Health and Welfare, 1999), an age-matched high activity man needs 2,950 kcal and 70 g of protein per day. The subjects consumed 1,422 kcal and 100 g protein more than the above recommended values.

Discussion

The decrease in TCR is one of the most characteristic features of OTS caused by an excess of training. We observed the pronounced decrease in serum testosterone concentrations (−35.0%) as well as the increase in plasma cortisol levels (+42.0%) after the training camp. Subsequently, TCR was dramatically decreased (−52.4%) compared with pre-camp values. In addition, the present subjects ran a mean of 35.5 km per day, almost twice as much as the distance of regular training. These results indicate that the subjects reached OTS during the 8-day severe training camp.

The most important finding of the present study is that the circulating blood leptin concentration is not affected by the changes in TCR during OTS in highly trained male distance runners. To our knowledge, this is the first study to investigate the effect of overtraining on plasma leptin concentrations together with both testosterone and cortisol levels in elite athletes. One explanation of why leptin levels did not show a significant change during the training camp is that the initial plasma leptin concentrations were extremely low in our subjects compared to age-matched normal healthy males (Nindl et al., 2002; Zafeiridis et al., 2003). Although significant changes in fat percentage were detected in this study, the subjects actually had quite a low absolute body fat mass (i.e., less than 2.0 kg) and low plasma leptin concentrations. Previous reports also suggested that the typical
characteristics of male endurance runners are low body fat mass and a low blood leptin level (Hickey et al., 1996; Sudi et al., 2001). In contrast, it is known that body fat mass is closely related to plasma leptin concentration in non-athletes (Considine et al., 1996; Koistinen et al., 1998). Taken together, it is assumed that the plasma leptin concentration is not necessarily dependent upon changes in fat percentage and/or body fat mass by exercise training, in well-trained distance runners.

In addition to body fat mass, NEFA has been shown to correlate with plasma leptin levels following acute or chronic exercise (Desgorces et al., 2004b; Dirlewanger et al., 1999; Duclos et al., 1999; Gomez-Merino et al., 2002; Karamouzis et al., 2002; Koistinen et al., 1998; Landt et al., 1997; Zaccaria et al., 2002). In many of the above-cited studies, subjects underwent exercise in a hypox-energy condition that was caused by large energy-consuming exercise or malnutrition. Subsequent low energy availability could lower the leptin level after exercise. In the present study, NEFA was not significantly increased after training, and Δ NEFA also had no significant relationship with Δ leptin (r = -277, ns). We infer that the unchanged levels of LBM, leptin and NEFA may be a reflection of sufficient energy intake during heavy training (Table 3), since leptin is involved in the control of energy homeostasis during the recovery from exercise (Desgorces et al., 2004a; Koistinen et al., 1998; Leal-Cerro et al., 1998; Olive and Miller, 2001).

While there was no significant correlation between Δ TCR and Δ plasma leptin, Δ serum testosterone was positively correlated with Δ plasma leptin. These results appear to be contradictory to a previous report of healthy men indicating that testosterone inhibits leptin secretion from adipose tissue and the serum leptin concentration is inversely correlated with the serum testosterone concentration (Luukkanen et al., 1998). However, another study showed that serum testosterone and plasma leptin levels decrease simultaneously after a 3-day fast but a leptin-analog supplementation prevents the decline of serum testosterone concentration (Chan et al., 2003). Therefore, it could be considered that leptin might regulate the serum testosterone level by joining the positive feedback control of the hypothalamic-pituitary-gonadal axis (HPGA) (Chan et al., 2003).

Many previous reports of healthy men indicate that the increase in serum cortisol concentration following acute exercise does not have an influence on the circulating leptin levels (Duclos et al., 1999; Essig et al., 2000; Koistinen et al., 1998; Zaferiridis et al., 2003). Moreover, it has been reported that, blood leptin concentration decreases after exercise training but serum cortisol levels are maintained in athletes (Gomez-Merino et al., 2002; Jürimäe et al., 2003). In the present study, there was no relationship between Δ serum cortisol level and Δ plasma leptin concentration. These findings suggest that cortisol may not play a major role in the regulation of the blood leptin level in the overtrained condition. In summary, we do not support the hypothesis that the decreased TCR observed during exercise training induces hyperleptinemia in highly trained male distance runners. Indeed, our data indicate that the plasma leptin concentration after the severe training camp is controlled independent of the changes in TCR and fat percentage in well-trained endurance athletes. Additionally, there is a possibility that plasma leptin concentrations modulate the secretion of testosterone in the overtraining condition. Further study is needed to elucidate the mechanisms regulating the blood leptin level in OTS.

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**References**


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