Leptin reduction after endurance races differing in duration and energy expenditure

Abstract Serum leptin concentrations are reduced in the presence of a negative energy balance. It has been demonstrated, however, that strenuous and prolonged exercise, which induces a marked negative energy balance, is not always followed by a reduction in serum leptin levels. We therefore analysed serum leptin concentrations before and after three endurance races, which differed in duration and energy expenditure (EE), with the aim of clarifying the relationship between the level of EE and the reduction in leptin levels. Forty-five males participated in one of three competitive endurance races, a half-marathon run [21.097 km, estimated EE 1,400 kcal (5,852 kJ)], a ski-alpinism race [about 45 km, estimated EE 5,000 kcal (20,900 kJ)], and an ultramarathon race [100 km, estimated EE 7,000 kcal (29,269 kJ)]. Blood samples for analysis of serum leptin, and plasma free fatty acids (FFA) were collected before and after the races. Pre-race leptin values were significantly correlated with both body mass index and body fat mass \( (r = 0.672 \text{ and } r = 0.699, \text{ respectively; } P < 0.0001) \). After exercise, serum leptin levels decreased significantly in the ultramarathon [from 4.15 (0.63) \( \mu g/l \) to 1.01 (0.15) \( \mu g/l \); \( P < 0.001 \)] and in the ski-alpinism race [from 1.10 (0.28) \( \mu g/l \) to 0.62 (0.15) \( \mu g/l \); \( P < 0.01 \)], but not in the half-marathon [from 1.38 (0.40) \( \mu g/l \) to 1.20 (0.36) \( \mu g/l \)]. Plasma FFA were found to have significantly increased in all three of the races, showing a negative correlation with the percent reduction in leptin \( (r = 0.369, P < 0.02) \). Our data indicate that only a prolonged endurance exercise involving a high EE can induce a marked reduction in circulating serum leptin levels.

Keywords Endurance exercise · Leptin · Energy expenditure · Free fatty acids

Introduction

Leptin, a protein that is expressed and secreted by adipose tissue, regulates food intake by acting as a hypothalamic signal of satiety. Under steady-state conditions of energy balance, leptin is considered a good index of the amount of adipose stores, while under non-steady-state conditions, leptin is no longer a reliable marker of body fat stores. In fact, a negative energy balance state, such as during prolonged fasting, decreases leptin concentrations, whereas a positive energy balance, such as during overfeeding, increases leptin levels (Kolaczynski et al. 1996).

Exercise is a physiological condition that effects a rapid increase of energy expenditure that is positively related to the intensity and duration of the exercise itself. Thus, a strenuous and prolonged exercise bout that induces a marked negative energy balance state would be expected to decrease leptin levels. However, reports on the effects of prolonged exercises on serum leptin concentrations have yielded contradictory findings. Serum leptin has been reported to be reduced or unchanged both after exercises performed in the laboratory, and after strenuous endurance races, such as marathons or ultramarathons (Hickey et al. 1996; Koistinen et al. 1998; Landt et al. 1997; Tuominen et al. 1997). These contradictory results may be due to differences in the experimental procedures used in the different studies (exercise protocols, diet before and during the exercise, circadian rhythm of leptin). We therefore analysed serum leptin concentrations before and after three competition endurance races that differed in duration and...
energy expenditure, with the aim of verifying the relationship between the level of energy expenditure and the reduction in serum levels of leptin.

**Methods**

Forty-five Caucasian males, all runners usually engaged in competitive races, voluntarily agreed to be included in the study and gave their written informed consent to participate. Each subject participated in one of three endurance competitive races. The energy expenditure for each race was estimated following methods described elsewhere (di Prampero et al. 1986).

The first race was a sea-level, flat half-marathon run (21.097 km), starting at 9.00 a.m., with a mean exercise time of 1 h 31 min and an estimated mean energy expenditure of 1,400 kcal (5.852 kJ).

The second race was a 45-km ski-alpinism race, alternating up-and-downhill running and cross-country skiing. Athletes started at 8:00 a.m. from 2,080 m, reached a maximum altitude of 4,226 m, and then went down to the finish at 1,637 m. The mean time of the race was 7 h 21 min, and the mean estimated energy expenditure was 5,000 kcal (20,900 kJ).

The third race was a 100-km ultramarathon race starting at 3:00 p.m., with a mean time of 15 h 27 min, and a mean estimated energy expenditure of 7,000 kcal (29,269 kJ).

All subjects were in a post-absorptive condition, having eaten a carbohydrate-rich meal about 2 h before the start of the race. During the race they drank water, and energy drinks (sweet tea, isotonic drinks), and ate energy bars ad libitum.

Before each race, in all subjects the body mass index (BMI) and body composition were assessed by bioelectrical impedance (BIA 101 RJL/Akern, Florence, Italy). Blood samples for measurement of serum leptin and plasma free fatty acids (FFA) were collected immediately before and within 20 min after the race, except in the ski-alpinism race, in which blood samples were collected from subjects in a post-absorptive condition the day before the race, at about the same time of day as the beginning of the race.

Serum leptin was measured using a specific double-antibody radioimmunoassay method (Sensitivity 0.03 μg/l; inter- and intra-assay coefficient of variability lower than 7.6% and 5%, respectively). Plasma FFA was determined by an enzymatic method (NEFA Quick “BMY”, Boehringer Mannheim Yamanouchi, Tokyo, Japan).

Statistical analysis

Data are expressed as mean (SEM). The statistical analysis of pre- and post-exercise values was made using the Wilcoxon test for paired data. Pearson’s correlation coefficient was used to test the relationship between leptin and the fat mass percentage and between the percentage of variation of leptin and FFA. The level of statistical significance was set at $P < 0.05$.

**Results**

The characteristics of the subjects under basal conditions are reported in Table 1: ski-alpinism subjects were the youngest, while the ultramarathon subjects were the oldest and had the highest BMI, body fat mass and resting leptin concentrations. The overall pre-race leptin values of subjects, were significantly correlated with BMI and with body fat mass ($r = 0.672$ and $r = 0.699$, respectively; $P < 0.0001$).

After exercise, serum leptin levels significantly decreased in subjects who participated in the two ultraendurance races [ultramarathon: from 4.15 (0.63) μg/l to 1.01 (0.15) μg/l; $P < 0.0005$; ski-alpinism race: from 1.10 (0.28) μg/l to 0.62 (0.15) μg/l; $P < 0.01$], while in the half-marathon, no significant reduction was found [from 1.38 (0.40) μg/l to 1.20 (0.36) μg/l; $P = 0.065$, Fig. 1]. The mean percent decrease in leptin was −71 (3)% in the ultramarathon, −40 (7)% in the ski-alpinism, and −2.5 (10)% in the half-marathon.

Plasma FFA levels significantly increased at the end of the races, with the absolute highest level being observed in the longest race [ultramarathon: from 471.3 (92.4) μmol/l to 1,801.4 (188.6) μmol/l, $P < 0.001$; ski-alpinism: from 486.9 (55.0) μmol/l to 1,484.0 (221.9) μmol/l, $P < 0.001$; half-marathon: from 733.8 (44.1) μmol/l to 1,234.8 (97.5) μmol/l, $P < 0.001$]. The percent variation in FFA showed a significant inverse relationship with the percent variation in leptin ($r = 0.369$, $P < 0.02$).

**Discussion**

The aim of our study was to investigate the effect on serum leptin concentration of three endurance races of different duration and energy expenditure. Two of the races, the ski-alpinism race and the ultramarathon, were very prolonged, medium- to high-intensity endurance races (7 h 15 min and 15 h 27 min, respectively),

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Half-marathon</th>
<th>Ski-alpinism</th>
<th>Ultramarathon</th>
</tr>
</thead>
<tbody>
<tr>
<td>Distance (km)</td>
<td>21.097</td>
<td>45</td>
<td>100</td>
</tr>
<tr>
<td>Mean [range] duration (h:min)</td>
<td>1:30 (1:08–1:50)</td>
<td>7:21 min (5:16–8:08)</td>
<td>15:27 min (13:00–17:00)</td>
</tr>
<tr>
<td>Energy expenditure (estimated kcal)</td>
<td>1,400</td>
<td>5,000</td>
<td>7,000</td>
</tr>
<tr>
<td>Number of subjects</td>
<td>23</td>
<td>11</td>
<td>11</td>
</tr>
<tr>
<td>Age (years)</td>
<td>44.3 (2.7)</td>
<td>34.6 (2.5)*</td>
<td>46.1 (3.2)*</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>23.2 (0.4)</td>
<td>22.0 (0.4)</td>
<td>26.4 (0.9)***</td>
</tr>
<tr>
<td>Body fat mass (%)</td>
<td>14.5 (0.7)</td>
<td>12.8 (0.9)</td>
<td>17.7 (0.8)***</td>
</tr>
</tbody>
</table>

* $P < 0.05$ vs ultramarathon and half-marathon;
** $P < 0.05$ vs half-marathon;
*** $P < 0.05$ vs ski-alpinism