

EFFECTS OF STRENUOUS TRAINING AND REPEATED CYCLES OF WEIGHT LOSS ON LEPTIN AND SEX HORMONES IN ELITE FEMALE WRESTLERS

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Abstract

Introduction: Subtle menstrual disturbances due to metabolic and hormonal alterations have been shown to affect a large proportion of elite female athletes. These alterations include leptin and sex hormones that have been shown to be related to body fat.

Aim of the study: We wished to determine if strenuous activity and repeated cycles of weight loss and regain affect leptin and other hormone levels in competitive female wrestlers.

Methods: Menstrual history, anthropometry, ovarian ultrasound scan, and hormone tests [leptin, oestradiol (E2), progesterone (P), luteinising hormone (LH), follicle-stimulating hormone (FSH)] were performed in a group of elite female wrestlers (A) and a group of aged-matched sedentary controls (C).

Results: Three athletes and no controls were oligomenorrheic (3-10 menses/yr). However the ovarian ultrasound scans were normal for all subjects. There was no significant difference between groups in age, height, or age at menarche. The correlation between body fat (%) and leptin was linear with regression coefficients ($r = 0.89$ (A) and $r = 0.83$ (C)) being similar in both groups. However, a significant difference was observed between the two groups for leptin levels ($P < 0.05$). E2 and P levels were within the normal physiological range in both groups ($E2 > 25 \text{ pg} \cdot \text{ml}^{-1}$; $P > 2.5 \text{ ng} \cdot \text{ml}^{-1}$) with P levels being lower for the athletes but the difference was not significant. Serum LH and FSH levels were also within the normal range ($> 1.5 \text{ IU} \cdot \text{L}^{-1}$).

Conclusions: No menstrual abnormalities were observed during the period of the study. Leptin levels were significantly lower for the wrestlers.

Key words: female, wrestling, weight loss, leptin, hormones

Introduction

Although wrestling is one of the oldest Olympic events dating back to the first ancient Olympic games in Greece, the participation of female wrestlers in high level competitive training is a recent phenomenon. Female wrestling was recognized by FILA (Federation Internationale des Luttistes Associees) in 1985 (1). Since then it has rapidly developed as a competitive sport and recently a growing number of females participate in international wre-

stling events, with Athens 2004 being their first Olympic games.

Competitive wrestling is performed in strict weight categories, and many “light” and “middle” weight wrestlers attempt to gain an advantage by competing at a weight just below the upper limit for their category. Drastically reducing food and water intake and exercising excessively is common practice among male wrestlers (2). Although the scientific data are not conclusive, the practice of “weight cutting”

(rapid weight loss), may alter hormonal status (3). Most recent reports suggest that the hypothalamic-pituitary-gonadal axis may be disturbed in male athletes (4). Reduced serum testosterone levels have been reported in male wrestlers both during training (preparation) and competition. No data exist for weight control behaviors of the elite group of women wrestlers. If they also practice "weight cutting" the same health and performance concerns apply to them as to their male counterparts (1).

Hypothalamic-pituitary gonadal function is disturbed in many female athletes involved in several other sports (5-6). Hypogonadism and restricted caloric intake are also established as significant factors in exercise-associated decreased bone density in women athletes (7) which can lead to increased incidence of musculoskeletal injuries (8) and premature osteoporosis (7). Although hypoestrogenic hypothalamic amenorrhoea in female athletes has been commonly associated with endurance training (9) women who are highly physically active could potentially be at risk of developing menstrual irregularities. Many studies have investigated the prevalence of menstrual irregularities amongst long distance runners (9), gymnasts, dancers (10), cyclists, triathletes (5), rowers, oarswomen (11), swimmers (12) and court sport players (6). From these it can be concluded that women participating in endurance sports such as distance running, cycling, and cross country skiing (7), as well as in sports which involve weight categories including horse racing, rowing, some martial arts, judo and wrestling are at greater risk than other athletes (7). For example, studies on female judo players have demonstrated that competitive practice can produce specific physiological effects on menstruation and bone metabolism (13, 11).

Several observations suggest a link between body fat and fertility (14). Addition-

nally, it has been recently reported that in females, maintenance of reduced body weight is associated with significant decline in plasma leptin levels (15). Leptin (leptos [Greek]: thin) is a possible candidate for mediation of the signal between fat stores and the reproductive system (16). As an adipocyte-derived protein, leptin acts in an endocrine fashion by reporting the size of the adipose tissue mass to hypothalamic leptin receptors. Anorexia nervosa patients have extremely low leptin levels and starvation is also associated with decreased immune function (17). Although the leptin receptor is also expressed in the human ovary, the mechanism by which leptin modulate fertility is not clear. Leptin has been reported to be very important in regulating the onset of puberty (17). Extremely thin women often stop ovulating (secondary amenorrhoea) and abnormally thin adolescent women enter puberty later (primary amenorrhoea) than their heavier counterparts, indicating that the fat tissue may produce a signal that regulates the reproduction (17).

The purpose of this study was to investigate the physiological and endocrinological effects of wrestling training and weight loss practice in women.

Materials and Methods

Subjects' background and training regime: Seventeen nulliparous healthy Greek women volunteered for this study. Nine subjects were aged 17-24 years and were competitive wrestlers and members of the Greek National team. Eight subjects were age-matched relatively sedentary control subjects (students of the University of Athens). Controls were not randomly selected in order to avoid any great difference between groups in age, weight and height at the time of the study; therefore the only variable between the groups that could affect hormone differences was poten-

tially the level of exercise and the weight cut practice of the wrestlers. Participants were informed of the purpose of the study and the procedures involved before giving their informed consent. All the experiments were carried out in accordance with the declaration of Helsinki.

The wrestlers ($n=9$) were participating in the international wrestling circuit, with a range of three to five years of international wrestling experience and a minimum of a national title and at least a fifth placing in a European or World Championship competition. During the study a pre-set training schedule (determined by the captain of the team) was the wrestlers' only activity. The training program was similar for each subject and composed of aerobic, anaerobic, weight and technical training, of moderate to high intensity. Physical activity for the control subjects was set at no more than three hours of aerobic exercise per week. The control subjects had never participated in competitive sports at representative level.

In judo it has been suggested that representative physiological characteristics and profiles are different and specific for each weight division, due to the constraints of weight loss and regain (11). Therefore, to partially overcome these limitations of heterogeneity, wrestlers belonging to the upper extreme weight class ($+75\text{kg}$ heavy-weight) were excluded. The groups were similar in habitual daily dietary energy intake for the duration of the study.

None of the participants were pregnant, lactating, taking hormonal or other medication or using anabolic steroids. Controls and wrestlers were of Caucasian descent and came from stable economic and social backgrounds.

Questionnaires' information: A questionnaire, adapted from a standard form developed by Brownell et al. (2), was completed by each subject. The first part of the

survey was designed to assess eating behavior and screen for eating disorders. The second set of questions was used to obtain the complete menstrual history of the participants, as well as to initially categorize each subject as having regular, normal length cycles (eumenorrhoeic), or being oligomenorrhoeic (3-10 menses/yr) or amenorrhoeic (<2 menses/yr). The last part of the questionnaire concerned physical activity. This was used to characterize each subject's past and present exercise habits. Three questions about bone fractures, current sports injuries, and cigarette smoking were also included. A Menstrual Distress Questionnaire (18) was also completed.

Body composition: The percentage of body fat was estimated according to the method of Durnin & Womersley (19). Four skinfold measures were taken from biceps, triceps, subscapular and supra-iliac regions, with a Harpenden skinfold caliper. The readings were taken on the right side of the body with the subject in the standing position, by the same experienced investigator.

Menstrual history, examination and ovarian scan: All the participants were advised to keep a menstrual diary for at least three cycles (one prior, one during and one after the study). The results of the hormone analyses were evaluated with reference to each subject's menstrual diary. Oestradiol (E2), progesterone (P), luteinising hormone (LH) and follicle-stimulating hormone (FSH) concentrations were analyzed during the menstrual cycle. Single ultrasound scan of the ovaries was also performed in order to detect any pathological situation. Control subjects were screened for a history of normal menstrual function.

Blood sampling: Venous blood samples were collected in ice-cold heparinised tubes and immediately centrifuged (3,000 rpm). The separated plasma was stored at -20°C prior to batch hormone analyses. Approximately two samples per week (the

same time of the day) were collected from each subject during the period of the study. Blood samples were collected post exercise from wrestlers and the controls. The hormone analyses were performed using solid phase chemiluminescent enzyme immunoassays (IMMULITE Euro DPC Ltd, Llanberis, UK). Total plasma leptin concentrations, were measured by RIA (21) The intra- and interassay coefficients and the sensitivity of measurement for leptin were 6.2%, 8.3%, and 0.5 ng/ml respectively.

Statistics: Mean values and standard error were calculated and data is reported as means $\pm$ standard error. Data were analysed using Excel Analysis tool (Descriptive Statistics, t-Test: Two-Sample Assuming Equal Variances). Correlation between % body fat and leptin were determined using simple linear regression analysis. Level of significance was set at $p < 0.05$.

Results

A comparison of the mean anthropometrical and physical characteristics of our two groups is shown in Table 1. None of the wrestlers were amenorrhoeic as assessed by the questionnaires. However, three athletes reported primary amenorrhoea

(delayed menarche). Those wrestlers had been competitive gymnasts earlier in their athletic career, with a history of menstrual dysfunction and dysmenorrhoea. During the period of the study, those subjects had menstrual cycles averaging 36-39 days in length, compared with the average 28-32 days of the other wrestlers and were classified as oligomenorrhoeic (Table 1). Duration of the menstrual cycle length in the controls ranged from 24-28 days. Evaluation of the single ultrasound scans of the ovaries revealed no abnormalities in any of the subjects which appeared to ovulate normally.

E2 and P levels were within the normal physiological range for perimenstrual, follicular and luteal phase (reference values for perimenstrual phase $E_2 = 25-55 \text{ pg} \cdot \text{ml}^{-1}$ & $P = 0.5-2.5 \text{ ng} \cdot \text{ml}^{-1}$, follicular phase $E_2 = 45-300 \text{ pg} \cdot \text{ml}^{-1}$ & $P = 0.5-1.5 \text{ ng} \cdot \text{ml}^{-1}$; luteal phase $E_2 = 55-180 \text{ pg} \cdot \text{ml}^{-1}$ $P = 2.5-19.5 \text{ ng} \cdot \text{ml}^{-1}$, Table 2). Serum LH and FSH were within reference values in both groups (perimenstrual phase & follicular phase $LH = 1.5-8 \text{ IU} \cdot \text{L}^{-1}$ & $FSH = 2.5-9.8 \text{ IU} \cdot \text{L}^{-1}$; luteal phase $LH = 1.5-6.5 \text{ IU} \cdot \text{L}^{-1}$ & $FSH = 1-9.8 \text{ IU} \cdot \text{L}^{-1}$, Table 2). Total leptin concentrations during the luteal phase were significantly lower ($p < 0.05$) in wrestlers com-

Table 1. Comparison table of mean anthropometrical findings and physical characteristics in female wrestlers (athletes), and sedentary women (control)

Parameters	Athletes (n=9)	Controls (n=8)
Age (yr.)	19.8 $\pm$ 1.0	23.8 $\pm$ 0.5
Age at menarche (yr.)	13.0 $\pm$ 0.3	11.7 $\pm$ 0.4
Eumenorrhoea [no. (%)]	6(66.6)	8(100)
Oligomenorrhoea [no. (%)]	3(33.3)	0(0)
Amenorrhoea [no. (%)]	0(0)	0(0)
Range of length of cycle (d)	28-39	24-28
Height (cm)	162.3 $\pm$ 1.7	163.5 $\pm$ 1.0
Weight (Kg)	51.6 $\pm$ 1.2	57.4 $\pm$ 1.5
Body fat (%)	18.6 $\pm$ 0.6	21.1 $\pm$ 0.9
Body mass index (kg/m ²)	19.6	21.6

Values are expressed as the mean $\pm$ SE

Table 2. *Hormonal status of female wrestlers and sedentary subjects*

Hormone/Phase	Athletes (n=9)	Controls (n=8)
E ₂ /Foll. (pg•ml ⁻¹)	128.9±1.5	152.6±2.7
E ₂ / P.M. (pg•ml ⁻¹)	25.0±2.8	28.8±2.9
E ₂ /Lut. (pg•ml ⁻¹)	82.5±5.1	69.5±3.4
P/Foll. (ng•ml ⁻¹)	0.95±0.2	2.29±1.9
P/Lut. (ng•ml ⁻¹)	7.5±4.4	6.8±3.1
LH/lut. (IU•L ⁻¹)	2.5±0.8	4.2±1.4
LH/Foll. (IU•L ⁻¹)	7.3±1.1	8.2±2.3
FSH/Lut. (IU•L ⁻¹)	3.4±0.5	6.1±1.8
LEPTIN (ng•ml ⁻¹)	11.6	17.5*

Foll.: Follicular phase, Lut.: Luteal phase

P.M.: perimenstrual phase

Values are mean ± SE

*Significantly different from controls (p < 0. 05)

pared to controls. (Fig.1 A). The correlation between % body fat and plasma leptin was calculated for both athletes and controls (Fig.1 B).

Discussion

Numerous methods are employed by male wrestlers to reduce bodyweight for competition, i.e. reduction of food intake (fasting and starvation), reduction of water intake, exercise to burn calories and induce sweating (metabolic dehydration) and thermal dehydration by having sauna (2). Female wrestlers in this study, reported at their interview that they lose weight rapidly before competitions (3-4 kg per 3-4 days) by a combination of sweating and excessive dietary and fluid restrictions, using the same methods as the male wrestlers. They also reported that they experience unusual delays in their menstrual cycle. From the data collected from the questionnaires it was evident that none of the wrestlers had a history of anorexia nervosa or exhibited behavior common to eating disorders, as have been reported for female recreational weight lifters and competitive body builders (20).

The anthropometrical characteristics of the wrestlers in this study were similar to

those in other reports on judo athletes (11, 13). Our study, indicates that the wrestlers (of the light and middle weight classes) may be labeled lean, although some of the subjects had a markedly higher mean percentage of body fat (20.2%) compared with endurance runners or ballet dancers (5, 6).

The hormonal findings indicate no menstrual abnormalities during the period of the study. Hormone levels are higher in the controls but not significantly different from those of the athletes group. The questionnaires, however, show that some of the wrestlers had irregular cycles and missed cycles altogether during their careers. Some of the wrestlers reported however that this happened, not during the normal training before the competition, but after strict dieting and strenuous exercise regime during and after the competition. The data in judo (11), suggests that there is a difference in judoist endocrinological findings monitored after a five-week training programme. As to their menstrual status, four of the 17 (23.5%) subjects had a history of oligomenorrhoea before the programme. Their number increased to nine (51.9%) at the end of the observation period (11).

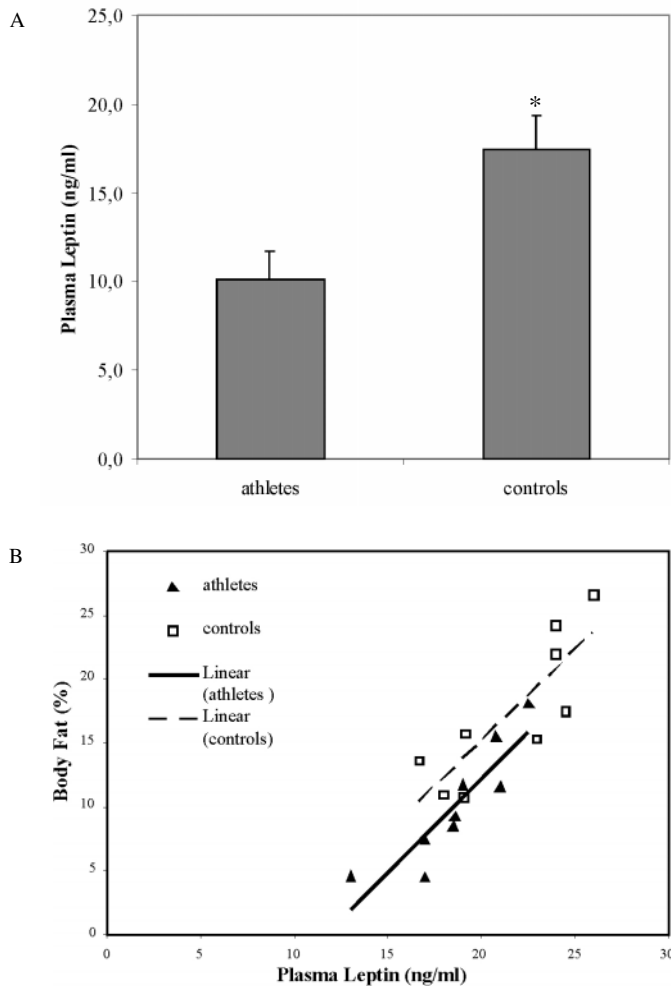


Figure 1. (A) Comparison of the means $\pm$ SE of plasma leptin levels (* $p < 0.05$). (B) Correlation between body fat and plasma leptin levels in athletes ($r = 0.89$) and controls ($r = 0.84$).

Dynamic weight loss and maintenance of a reduced body weight is associated with a significant decline in plasma leptin as has previously reported (22) and also observed in this study where total leptin concentrations were significantly lower ($p < 0.05$) in wrestlers compared to controls. However the leptin levels were normal and previously reported relationship between circulating leptin (23-25) and fat context were maintained for both groups.

Although the mechanism by which leptin modulates fertility is not clear, there are indication that leptin modulates the repro-

ductive endocrine system in the hypothalamus, pituitary, and ovary (14, 26, 27). The present study, examines the effects of competitive wrestling on the hormonal levels of competitive female wrestlers. Elite female wrestlers practice "weight cutting" like male wrestlers. However, exercise training as previously reported appears to have a greater effect on systemic leptin levels in females than in males (15, 22). Food restriction, fasting and various dehydration methods are the common weight loss techniques. The authors strongly oppose the application of those techniques in female

wrestling practice. The hormonal findings and especially the reduced levels of leptin, can be interpreted as an early indication of a potential future metabolic imbalance. Although the trend of lower leptin levels observed is not alarming, it should be evaluated with further research and monitoring of female athletes that perform weight loss and can be part of a routine medical examination protocol.

Practical application: To date, wrestling is a new female event and it is essential for educators, health professionals, dieticians, exercise scientists and wrestlers to make a cooperative effort to systematically collect data on the body composition, menstrual status, energy and nutritional demands, weight control behavior, growth and maturation of the selected group of female athletes. International regulations involving weight categories and major competitions should consider those observations. The differences of the female physiology and reproductive system require a different approach to training and competition. This is a call for further focusing in a developing event.

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