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Original Research

## Leptin Fluctuations in Trained Horses, During a Work Season



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## ABSTRACT

The effect of long-term training on plasma leptin concentration in horses remains unknown. The aim of the study was to evaluate the influence of training on plasma leptin concentration in race and sport horses, during a work season. Thirty-four clinically healthy horses were assessed during their race and equestrian training. They were 12 adult sport, saddle-breed stallions (group S), 12 naïve, 3-year-old Purebred Arabian horses involved in race training (group R1), and 10 4-year-old Purebred Arabian horses, which were continuing their race career (group R2). Blood samples were collected at rest, four times during the training season, and from March to September. Plasma leptin and cortisol concentrations were measured and analyzed. In group R1, the mean value of leptin gradually decreased from March to September (from  $10.8 \pm 7.4$  ng/mL to  $1.1 \pm 0.4$  ng/mL), whereas the plasma cortisol concentration was low and unchangeable during the study ( $63.2 \pm 16.1$  ng/mL). In group R2, plasma leptin concentration was generally below 1.0 ng/mL despite the fact that the cortisol concentration increased during the study, reaching the mean value of  $656 \pm 693$  ng/mL in September. In group S, the studied parameters did not exceed the values of  $1.90 \pm 0.22$  ng/mL for leptin and  $75.0 \pm 21.2$  ng/mL for cortisol. The values of coefficient correlation for studied parameters were statistically insignificant. Thus, the plasma leptin concentration is not dependent on cortisol concentration in trained horses. The training routine leads to decrease in the plasma leptin concentration in young horses.

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## 1. Introduction

In horses, as in other mammals, leptin is a peptide hormone synthesized mainly in adipocytes and secreted by adipose tissue. Therefore, a circulating leptin level corresponds positively with body fat mass [1–4] and the age of a horse [5,6]. Horses do exist, though, which have high or low resting leptin levels [2,3,7]. Aside from the body fat mass or body condition score (BCS), plasma leptin concentration can transiently increase after a meal and decrease with

negative energy balance [8–11]. The fluctuations in plasma leptin levels are associated with the physiological role played by this hormone. Namely, leptin decreases appetite and food intake and increases energy expenditure [1,12–14]. Hence, the regulation of circulating leptin concentration and the influence of leptin on metabolism is very important for the homeostasis, especially in the case of athletic horses. Moreover, the plasma leptin concentration is associated with the occurrence of some behavioral and somatic disorders, such as crib biting, insulin resistance, and mortality in foals [15–18].

It is known that leptin synthesis and release are stimulated by corticosteroids and insulin, whereas adrenaline plays an opposite role [19,20]. Moreover, the *in vitro* reports found only a small stimulatory effect of insulin on

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leptin release as compared to the market increase in leptin synthesis and release due to corticosteroids [19]. It is also known that cortisol and adrenaline secretions increase during exercise, whereas the insulin level decreases. The ratio of changes in the levels of these hormones depends on the duration and intensity of the performed exercise as well as on the horse performance [5,21,22]. Thus, exercise changes the endocrine profile, but the final effect of exercise on the plasma leptin concentration remains unclear [23]. Furthermore, the effect of long-lasting training on plasma leptin concentration remains unknown. Carter et al [24] did not observe any effect of moderate exercise training in overweight, insulin-resistant horses. Kędzierski and Kapica [25] reported that plasma leptin concentration in young Standardbred horses was higher in winter than in summer. A similar effect was described in trained Iberian show horses; however, Amato et al [26] pointed out the influence of intense exercise performed by the horses during summer show periods. The opposite effect was noted in untrained, pastured mares and female donkeys, in which the plasma leptin concentration was lower in winter than in summer [6,27]. These observations suggest that training changes plasma leptin level in athletic horses, in comparison to the plasma leptin levels in untrained, pastured equids. Therefore, the aim of the study was to test the hypothesis that long-lasting training decreases the plasma leptin concentration in horses. To test this hypothesis, young Purebred Arabian horses during their race-training season were studied and compared to adult horses used in equestrian sports. These adults were the control group in this study. The fluctuations in the plasma leptin concentration were analyzed together with changes in the plasma cortisol concentration because cortisol is the main regulator of leptin synthesis in regularly exercised horses.

## 2. Materials and Methods

### 2.1. Horses

Thirty-four clinically healthy horses were assessed during their race and equestrian training. Initially, the number of horses included in the study was larger; however, some horses had to be excluded because of injuries or because they were transferred to another training place. The horses used in the study completed their routine training. They were 12 sport stallions (six dressage and six jumping) that were 5–16 years old. These were warmblood horses bred for classical equestrian sports (group S). There were also 12 naïve, 3-year-old Purebred Arabian horses (six mares and six stallions) which were included in race training (group R1); and 10 4-year-old Purebred Arabian horses (four stallions and six mares), which had previously training on the race track and were continuing their racing career (group R2). The horses from group S were used in the study as a control group because of their experience, low-intense exercise, and because they were adults. All studied horses were trained and treated in manner characteristic for their use in sport or races. The study was performed between March and September of one training season. In the winter preceding the study, the horses had not been regularly trained. They had access to paddock a

few hours a day, if weather permitted. The sport horses spent the winter in their training center. They were housed in two stables under comparable social and environmental conditions. Each horse was kept in a box stall measuring 4 m × 4 m and lined with straw bedding. In addition, the horses from group S were walked on an automated horse walker once a day and were ridden recreationally inside a riding arena 3 or 4 days a week. The race horses spent winter in their studs. They were housed in individual boxes measuring 3.5 m × 4 m.

In late March and early April, the horses started to be regularly trained. Dressage horses were put through a technical training of increasing difficulty within a standard arena 1 hour a day. Jumping horses routinely overcame 30 to 55 obstacles which were of maximum high 1.3 m (in April) to 1.5 m (in July and September) during daily training sessions. From May, the horses participated in shows and competitions about once a month. The Purebred Arabian horses from group R1 were trained in their stud. It was the first training and racing season in their life. The training sessions were performed for about 1 hour a day, 4 to 5 days a week. The riders rode the horses at a walk and trot for approximately 10 minutes as a warm-up exercise. Then, the horses cantered or galloped on the sand track. The speed and duration were individually adapted to the level of each horse's performance. After the exercise, the horses were put on an automatic horse walker for 45 minutes. In addition, 1 or 2 days a week, the horses were ridden in the woods for relaxation. From June, the horses participated in official races. This meant the horses were regularly transported to the race track at least once a month. The horses from group R2 were trained at the race track (Sluzewiec, Warsaw, Poland). They were brought from their studs about 2 months earlier to acclimatize to the new conditions and begin the training. They were housed in one stable and were trained by the same trainer. Their training routine consisted of gallops on the sand track 6 days a week. At the end of May, they started to compete in official races at least once a month. During the study, each horse received an individual ration of hay, oats, and fed concentrate, according to their body condition and workload. The feed dose was split into three feedings: 06:00 hours, 12:00 hours, and 18:00 hours. For example, feeding regiment (kg/horse) for R1 horses in July was as following: 5.5 oats, 6.0 meadow hay, and 0.8 fed concentrate. A mineral salt block and fresh water were made available *ad libitum*. For each horse, the BCS was always determined by the same, experienced veterinarian. The rating system devised by Henneke et al [28] was used. Scores ranged from 1 to 9. All horses from groups R1 and R2 took part in official races and were handicapped on the end of the race season. The officially published handicap value for each horse was divided by the mean handicap for all horses on the same age which had started on the race track during whole season (relative handicap).

### 2.2. Blood Sampling and Analysis

Blood samples were collected four times during the training season. Collections were done in 2-month intervals, in the middle of March, May, July, and September.

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