# Hormonal and biochemical profile in elite sportsmen during the preparation season

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Abstract. Concerns have been raised regarding the effects of short time intensive training on adolescent athletes. The aim of the present study was to investigate six weeks of volleyball training on hormonal and biochemical profile in elite high school male volleyball players in Golestan province (Iran) in preparation season to take part in global competitions of champion school in France. Subjects of training group consisted of 12 elite volleyball players in premier league students and the control group consisted of 12 healthy and non-athlete boy students. The training group practiced at 60-85% maximum heart rate (MHR) for 6 weeks, 90 minutes per session, 6 sessions per week and 3 sessions a day (first session: technical practice, second session: tactical practices and third session: technical/tactical practices). Blood samples were taken following 12 hours of fasting both before and after the volleyball training. cortisol and lactate dehydrogenase concentrations significantly decreased in the training group following 6 weeks of Preparation training. With the exception of cortisol there was no significant difference between the groups regarding lactate dehydrogenase variable. Glucose concentration significantly increased in the training group following the 6 weeks Preparation training, also significant difference was observed between the control and training group. Testosterone, insulin and insulin resistance levels were increased and cortisol/testosterone ratio was decreased in the training subjects following the 6 weeks of Preparation training (P>0.05). However, the results revealed no significant difference between the two groups regarding these variables. Our results show that volleyball training during the 6 weeks in the preparation season can lead to abnormalities in indices of glucose homeostasis. Also the results suggest that along with the improvement of anaerobic and aerobic characteristics, training reduces the catabolic and inflammatory response to exercise.

**Keywords.** Cortisol, glucose, insulin, insulin resistance, testosterone, volleyball player

# Introduction

In volleyball, the first part of the season is a period of heavy training loads that aim to develop technical and tactical skills, as well as achieve adequate physical fitness for the competition period (Mielgo-

Ayuso et al., 2013). Nowadays, because students' matches occur at the school season there is a limited time for preparing and taking part in matches. For this purpose, preparation training performs with high volume, intensive and very short-term. This imposed high physical and psychological pressure on the players (Gharakhanlou et al., 2002) that caused changes in some hormonal (e.g. cortisol; C, testosterone; T and insulin; I) and biochemical variables such as glucose homeostasis and some factors involved in inflammation and pressure (e.g. lactate dehydrogenase). It is difficult to establish the effects of training on some hormonal and biochemical profile of professional volleyball players. This is because, apart from the personal characteristics of each player, particular features of their training, especially those focused on competition (Tambalis et al., 2009) can substantially modify hormonal and biochemical profile. Ruiz et al. (2004) commented that volleyball is a sport with a strong component of physical stress; however Hormonal and metabolic changes during volleyball drills mainly depend on the duration and intensity of exercise. It has been shown that an increase in the amounts of circulating plasma cortisol or a decrease in testosterone can result in whole-body insulin resistance (Mendes et al., 1985). In men's team athletics, competition is usually associated with an increase in testosterone level and, depending on the sport, an increase in cortisol (Edwards & O'Neal, 2009). Thus, the primary aim of this study was to evaluate potential changes in the some anabolic (testosterone) and catabolic (cortisol) hormonal that might be induced by 6 weeks of training in elite high school male volleyball players. Very few studies have examined the effect of training on the glucose homeostasis indexes (insulin, glucose and insulin resistence) and inflammatory and pressure markers in adolescent elite athletes and in team sports, which are very popular in these age groups. We hypothesized that, as seen previously in individualized sports, volleyball practice will lead to a significant desirable change in both hormonal and biochemical profile. Very few studies examined the effect of team sports training on these variables. Therefore, the secondary aim of this study was to monitor the physiological stress placed upon players

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during 6 typical week of a season on responses of new biomarkers of Muscle injury (lactate dehydrogenase; LDH).

## Methods

#### Subjects

The study population consisted of male students aged 16-18 years, studying at the High Schools in Golestan Province, Iran. A quasi-experimental method with preand post-test approach was implemented, as well as training and a control group. The sample size of the training group was 12 elite high school male volleyball players in Golestan province. They were the first rank in students' volleyball games and will play as Iran team in championship games in France. They were chosen by random sampling method. Also 12 healthy high school students that are not players were randomly chosen as control group (Table 1).

Table 1. Demographic characteristics of subjects.

| Variables                              | Control group<br>(n=12)            | Training group<br>(n=12)             |
|----------------------------------------|------------------------------------|--------------------------------------|
| Age (yrs) Height (Cm)                  | $17.08 \pm 0.66$ $190.33 \pm 6.02$ | $17.00 \pm 0.73$ $182.58 \pm 10.24$  |
| Weight (Kg)<br>Body Mass Index (kg/m²) | $65.12 \pm 4.52$ $18.14 \pm 0.39$  | $64.12 \pm 4.82$<br>$19.39 \pm 0.87$ |

Subject of the two test groups were similar in terms of age, height, weight and BMI (P> 0.05).

The data are illustrated as mean  $\pm$  SD.

After getting permission from department of education in Golestan province and receiving informed consents from students and their parents Researchers entered training camp. Body weight (BW) was measured in kilograms using a SECA scale, to the nearest 0.1 kg., and height using a stadiometer to the nearest 0.5 cm. Body mass index (BMI) was then calculated using the formula BW/height2 (kg/m2).All players were on a balanced diet supervised by a dietician throughout the study. The study protocol was approved by the local Ethics Committee. The experimental procedures, associated risks, and benefits were explained to eligible players before they gave written informed consent to participate. The training group took part in preparation training for participating in global matches of school championship in France. The training group practiced for 6 weeks, 6 days a week, 3 sessions a day (first session: technical practice, second session: tactical practices and third session: technical/tactical practices) for 90 minutes per session. The first week they practiced with intensity of 50-60% Maximum heart Rate (MHR), the second week with 60-65% MHR, the third week with 70-75% MHR, the fourth week with 80-85% MHR, the fifth week with 70-75% MHR and the sixth week with 60-65% MHR. Training sessions were at 8-9.30 and 10.30-12 A.M and

16-18 P.M. In practice sessions the subjects first did warm up for 10 minutes and then after the main volleyball training they did 15 minutes cool dawn exercise to recovery.MHR was measured using the following equation: MHR=206-(0.88xage), in order to adjust the training intensity and while-training heart rate (Gulati et al., 2010).MHR was calculated for all the subjects and then they were instructed to do training at their 60-85% MHR. The control group, however, were asked not to do any exercise training during the training program.

#### Blood collection and analysis

Venous blood samples were drawn after 12 hours of fasting from the ante-cubital fossa of the forearm, between 8.00 and 9.00 A.M. None of the players trained the day before the samples were taken. Then the lactate dehydrogenase as a marker of cell injury was measured with photometer 5010 device made in Germany. Total testosterone and cortisol, were quantified on the ELISA analyzer with the use of commercially available ELISA kits (Diagnostic Systems Laboratories, Inc., Webster, TX, USA). Blood glucose was measured using the enzymatic colorimetric method (Glucose, Colorimetric Enzymatic, Pars Azmun, Tehran, Iran). Serum insulin levels were determined by sandwich ELISA (Insulin, ELISA, DRG, Marburg, Germany). Insulin resistance was measured by Homeostatic Model Assessment (HOMA-IR) (Matthews et al., 1985) using the following equation:

HOMA-IR = fasting glucose [mmol/l] x fasting insulin  $[\mu U/ml]/22.5$ 

#### Statistical analysis

Statistical analyses were performed using the IBM Statistical Package of Social Sciences (IBM SPSS Statistics, v. 20.0 for WINDOWS). Kolmogorov-Smirnov tests were used to analyze for normal distribution. All data are reported as means ± standard deviations. Paired and independent t-tests were used for within-group and between-group comparisons, respectively. The level of statistical significance was set at P≤0.05.

#### Results

The results showed no significant difference in weight, age, height and BMI between the control and experimental subjects (P>0.05) (Table1). After six weeks of preparation, the amount of BMI significantly decreased in training group (P<0.05) also there was a significant difference between the two groups in this variable.

Cortisol and lactate dehydrogenase concentrations significantly decreased in the training group following 6 weeks of Preparation training (P<0.05). With the exception of cortisol there was no significant difference between the groups regarding lactate dehydrogenase

variable. Glucose concentration significantly increased in the training group following the 6 weeks Preparation training, also significant difference was observed between the control and training group. Testosterone, insulin and insulin resistance levels were increased and cortisol/testosterone ratio was decreased in the training subjects following the 6 weeks of Preparation training (P>0.05). However, the results revealed no significant difference between the two groups regarding these variables. Cortisol, testosterone, cortisol/testosterone ratio, insulin, glucose and insulin resistance levels decreased and lactate dehydrogenase increased none significantly in control group (Table 2).

Table 2. Mean and standard deviation of the research variables before and after 6 weeks of preparation training.

| Variables                      | Groups   | Before training    | After training     | Sig.<br>(dependent t) | Sig.<br>(independent t) |
|--------------------------------|----------|--------------------|--------------------|-----------------------|-------------------------|
| BMI (kg/m²)                    | Control  | $19.52 \pm 3.03$   | $19.39 \pm 3.02$   | 0.075                 | 0.000*                  |
|                                | Training | $17.85 \pm 1.32$   | $18.14 \pm 1.35$   | 0.000*                |                         |
| Cortisol (micg.dl)             | Control  | 128.97 ± 16.50     | $118.68 \pm 9.06$  | 0.597                 | 0.717                   |
|                                | Training | $161.52 \pm 4.69$  | $143.93 \pm 7.13$  | 0.015*                |                         |
| Testosterone (ng/ml)           | Control  | $4.75 \pm 0.54$    | $4.68 \pm 0.59$    | 0.754                 | 0.810                   |
|                                | Training | $5.27 \pm 0.18$    | $5.35 \pm 0.19$    | 0.903                 |                         |
| Cortisol/ Testosterone (Ratio) | Control  | $31.58 \pm 16.56$  | 29.55 ± 12.57      | 0.644                 | 0.784                   |
|                                | Training | $30.82 \pm 3.17$   | $27.44 \pm 6.27$   | 0.147                 |                         |
| Glucose (mmol/l)               | Control  | 77.75 ± 5.17       | $77.25 \pm 6.91$   | 0.665                 | 0.000*                  |
|                                | Training | $72.16 \pm 2.32$   | $85.75 \pm 5.89$   | 0.000*                |                         |
| Insulin (µU/ml)                | Control  | $8.78 \pm 1.44$    | $7.93 \pm 1.32$    | 0.871                 | .351                    |
|                                | Training | $7.25 \pm 1.00$    | $7.57 \pm 1.79$    | 0.520                 |                         |
| Insulin resistance (HOMA-IR)   | Control  | $30.34 \pm 17.52$  | $26.93 \pm 14.96$  | 0.450                 | 0.321                   |
|                                | Training | $23.55 \pm 11.74$  | $28.55 \pm 23.03$  | 0.491                 |                         |
| Lactate dehydrogenase (u/l)    | Control  | $304.16 \pm 11.00$ | $328.83 \pm 4.94$  | 0.059                 | 0.000*                  |
|                                | Training | $395.66 \pm 18.64$ | $338.00 \pm 18.49$ | 0.003*                |                         |

<sup>\*</sup> Statistical significance (P≤0.05)

#### Discussion

The key findings from our study are; 1) A significant reduction in BMI; 2) A significant increase in glucose; 3) significant decrease in cortisol and LDH levels and 4) significant changes in LDH, BMI and glucose between the two groups after 6 week of volleyball training.

In contrast, Edwards et al. (2007) stated that Women's intercollegiate volleyball lead to levels of cortisol (C) and testosterone (T) increased when women participated in athletic competition. Levels of C and T appear to rise in parallel during competition and increases in levels of one hormone are significantly related to increases in the other. Consistent with the results, Eliakim et al. (2009) demonstrated that volleyball practice resulted in a significantly reduced cortisol response (cortisol:  $4.2 \pm 13.7$ vs.  $-4.4 \pm 12.3$  ng · ml), before and after training, respectively; p < 0.02). Li et al. (2012) reported However, the cortisol concentrations in volleyball players were markedly higher compared with those in sedentary controls. Moreira et al. (2013) results with elite young volleyball players showed that match intensity, cortisol concentration were affected by the match importance. These results indicate that monitoring C responses, in conjunction, during training and competition, would provide valuable information regarding how athletes cope with sports induced stress. This study provided knowledge about the effect of match importance on markers related to stress that may help coaches to avoid excessive training loads reducing the likelihood to decrements on mucosal immunity and its consequent risk to upper respiratory tract infections, which in turn might

affect the performance. Edwards & Kurlander (2010) have shown cortisol levels were relatively unchanged during warm-up, but typically rose during volleyball competition. Thus, as women prepare for athletic competition by warming up, testosterone levels rise in apparent anticipation of the coming contest and then remain high through the period of play. In volleyball, after-practice testosterone level was significantly higher than before-practice level, and practice session increases in testosterone (but not cortisol) were positively correlated with increases in testosterone during intercollegiate competition. When practice competitive play share as yet undetermined key elements, individual differences in this endocrine response to "competition" appear stable across practice and intercollegiate competition. Eliakim et al. (2013) stated that volleyball practice led to significant increases in testosterone in men. Exercise had no significant effect on cortisol levels. These changes may serve as an objective quantitative tool to monitor training intensity in unique occasions in team sports. Bouassida et al. (2009) demonstrated that plasma insulin decreased (p < 0.01) significantly during short and prolonged sub-maximal cycling exercise protocols (in volleyball players) this hormone remained lower. There was no significant change in corisol and glucose during and after both protocols for all subjects (Eliakim et al., 2009). Kuo et al. (2006) in trained and competition states, among sixteen highly trained volleyball players stated that insulin levels were significantly elevated by competition states above trained states. Plasma cortisol in the competition states were significantly elevated above trained states, while testosterone levels and C/T were not significantly

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different between two states (Kuo et al., 2006). Inconsistent with this study, Ghasemnian et al. (2013) reported that after eight weeks of interval endurance training, insulin resistance decreased, in overweight and obese adolescents. It has been shown that an increase in the amounts of circulating plasma cortisol or a decrease in testosterone can result in whole-body insulin resistance (Mendes et al., 1985). In men's team athletics, competition is usually associated with an increase in testosterone level and, depending on the sport, an increase in cortisol (Edwards & O'Neal, 2009).

Hormonal responses to exercise depend on some factors such as duration, type, and intensity of exercise, genetic background, gender, nutrition, age, circadian pattern of secretion and the participant's fitness. The amount of muscle mass involved in the activity, intensity and volume of exercise, diet, age and Practice are some factors that influence testosterone. It has been shown that intensive exercise protocols that involve multiple joints and muscle building increases acute testosterone concentrations (Sourati Jabloo et al., 2012). Increase in T level, may be due to an increase in stimulates the adrenal, stimulatory effect of lactate or testosterone secretion ability to adapt. The researchers reported that testosterone response to exercise training in trained subject in compare with untrained subject is higher. Therefore, elite volleyball player participator may explain the rise in testosterone levels. Reduction in LDH level indicates that this period of training did not lead to cell injury. Likely duration, intensity and volume of exercise have been sufficient.

In sum, the results suggest that along with the improvement of anaerobic and aerobic characteristics, training reduces the catabolic and inflammatory response to exercise. However, preparation training for optimizing the profile of some metabolites and indices of glucose homeostasis has not been enough.

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