

Original empirical article

**THE EFFECT OF DIFFERENT INTENSITY OF EXERCISE ON  
SALIVARY STEROIDS CONCENTRATION IN  
ELITE FEMALE SWIMMERS**

UDC 547.92:797.015

**B. Yazdanparast<sup>1</sup>, M.A. Azarbayjani<sup>2</sup>, M.J. Rasaei<sup>3</sup>,  
Morteza Jourkesh<sup>4</sup>, Sergej M. Ostojic<sup>5</sup>**

<sup>1</sup>IA University, Varamin Branch, Iran,

<sup>2</sup>Exercise Physiology Dept., Faculty of Physical Education, IA University,  
Central Tehran Branch, Tehran, Iran;

<sup>3</sup>Dept. of Biochemistry, School of Medical Sciences, Tarbiat Modares University, Tehran, Iran;

<sup>4</sup>Department of Physical Education and Sports Science, IA University, Shabestar Branch, Iran;

<sup>5</sup>Faculty of Sport and Tourism, PA University of Novi Sad, Serbia

**Abstract.** *The purpose of this study was examine to effect of different exercise intensity (low, medium and high) on the salivary concentration of Cortisol and DHEA in elite female swimmers. 10 young female swimmers voluntarily agreed to be the subjects. The subjects participated in a 6-week exercise program. The exercise program was divided into three time segments: the basic endurance session during the first two weeks, special endurance session during the second two weeks and hard exercise session during the last two weeks. Saliva samples were collected at the end of each two weeks before and immediately after exercise from the study group. Concentrations of salivary steroids were determined by using the ELISA method. The findings of the research show that: exercise with low, medium and high intensity does not have a significant effect on the concentration of cortisol, DHEA and the ratio of salivary DHEA:C of elite girl swimmers,  $p>0.05$ . There is no significant correlation between concentrations with salivary cortisol after exercise with high and low intensity,  $p>0.05$ . But a negative significant correlation was found between concentrations of salivary cortisol after exercise with medium intensity,  $p<0.05$ . There is no significant correlation between concentrations with the ratio of salivary DHEA:C after exercise with low, medium and high intensity,  $p>0.05$ . In conclusion, the results of this study show that salivary steroids are affected by the intensity of the exercise.*

**Key words:** *Cortisol, DHEA, Saliva, Swimmer, Training*

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Received March 21, 2009 / Accepted May 11, 2009

**Corresponding author:** Morteza Jourkesh, MSc

Department of Physical Education and Sports Science; IA University, Shabestar Branch, Iran

Tel: (+98)-9143124670 • E-mail: jourkesh\_2006@yahoo.com; mjourkesh@iaushab.ac.ir

## INTRODUCTION

In women, nearly 90% of circulating T derives from the metabolism of peripheral precursors and in particular from DHEA. DHEA is a metabolic intermediate in the pathway for the synthesis of T, estrone and estradiol (Filaire et al., 1998).

Cortisol (c) and testosterone (T) and DHEA have been recommended as good markers of training stress. Indeed, the balance between catabolic (c) and anabolic hormones (T, DHEA) may have important implications for performance and recovery processes.

The changes in DHEA and cortisol hormones are related to physiology activity parameters such as intensity and length of training and the area of training condition.

In the case of swimming, data concerning C and T responses to performance are conflicting. Mujika et al. found that increases in the T/C ratio were positively correlated with improvement in swimming performance. Copasalmi et al. found that an increase in the cortisol in anaerobic training causes an increase in its density (Chatard et al., 2002; Filaire et al., 1998; Guyton & John, 1996). The (DHEA):(C) ratio is a training index of female athletes (Filaire et al., 1998). This ratio is influenced by the intensity and length of training and can be a sign of pressure due to the training.

Beaven et al. completed a study involving female and male soccer players and found that cortisol and testosterone concentration in both groups increased (Dimitriou et al., 2002). O'Connor et al. studied different salivary cortisol levels in swimmers during the extensive activity session and found that there was no significant change in type (O'Connor et al., 1998). Filaire et al. found no significant differences in the salivary cortisol levels between 8 AM and the following 24 hours in the swimmer and control group, but this level among handball players was much higher than in the other two groups (Filaire et al., 1998). Jacks et al. observed that during the most intensive training, the levels of salivary cortisol were higher than during other times. A significant difference in the salivary cortisol levels was not found during breaks and less intensive training (Jacks et al., 2002). Chatard et al. found that there was no significant relation between the cortisol and DHEA levels and performance (Chatard et al., 2002). Filaire et al. observed that salivary DHEA levels were higher among athletes, with an increase in the salivary DHEA levels during competition. There were no significant differences in the cortisol levels among the athletes during the 16th week of training, while the DHEA C ratio in all female athletes was 30% higher than during the first week (Filaire et al., 1998).

In the case of swimming, few data are available on DHEA or DHEA:C responses to performance and training. As for T:C ratio, an altered DHEA:C ratio may influence the anabolic/catabolic balance. Indeed, the DHEA mode of action involves its transformation in active sex steroids, promoting overall anabolic effects in several tissues (Chatard et al., 2002).

Therefore, the aim of this study was to investigate whether there was a relationship between C, DHEA, and training intensity in elite female swimmers. It is possible that individual differences between hormones, performance and swimming training may obscure any relationship when only group data are present.

## METHOD

### Participants

Some of the physiological characteristics of the 10 young female swimmers who voluntarily agreed to be our subjects are shown in Table 1. The subjects read and signed an informed consent form with a medical history questionnaire before the beginning of the study. Then, the nature of the study was explained to them and they were given an opportunity to ask questions about anything that was unclear.

None of the persons have a previous history of hormonal disorders and for the duration of the research, none of them were undergoing therapy. All of the subjects participated in all of the exercise sessions.

Table1. Descriptive characteristics of the subjects

Age.yr	13.3 ±2.35*
Height.cm	152.3±11.41
Weight.kg	48.7± 14.22
Body fat,%	15.93±0.6
BMI. kg/m <sup>2</sup>	20/53±3/92
Vo <sub>2</sub> max.ml.kg.min	
Anaerobic threshold b.m	173.8 ± 10. 45

\*Results are expressed as mean± standard deviation

### Training Protocol

The subjects participated in a 6-week exercise program, during which time all of the exercise sessions performed between 5 and 7 p.m., three times per week. The exercise program was divided into three time segments:

- 1) Base endurance session: during this session, the intensity of exercise was low, which means that the selected participants swam at around 60 to 70% of their best records.

Their heart beats were 140-150 beat/minute, the type of exercise was endurance of 1, and the speed was supplied by the phosphagen system.

- 2) The specific endurance session: in this session, the selected participants swam at around 71 to 80% of their best records. The intensity of the exercise was moderate, their heart beats were 160-170 beat/minute, and type of exercise was endurance of 2.

The speed was limited to lactic acid production.

- 3) The severe exercise session: the intensity of exercise was high in this session. The selected participants swam at around 81 to 95% of their best record. Their heart beat was in the threshold of the non-aerobic. The type of exercise was endurance of 3, the speed was limited to bearing lactic acid.

### Saliva collection

The subjects provided samples of saliva during W2, W4 and W6 when exercise intensity was low, moderate and high without stimulation. They spat directly into a plastic tube before and after the exercise. The samples of saliva were stored in a freezer at -30°C.

### Hormonal assay

The Cortisol concentration and saliva DHEA were specified using the ELISA method. Saliva cortisol concentration was specified with an accuracy of -5ng/ml by means of a commercial kit of ELISA that was produced by the Italian company of RADIM, and saliva DHEA concentration was specified with an accuracy of 0.08 mic/ml by means of a commercially indicated measurement of descriptive characteristics.

**Procedure**

Means and standard deviations were used to describe the quantitative variables.

The measurements of salivary cortisol and DHEA-S were tested for effects of "time" and "intensity" in a repeated-measures analysis of variance (RM-ANOVA). Before these analyses were performed, the frequency distributions were tested for normality using the Kolmogorof-Smirnov test. Data in text and figures are given as the mean ± st.dev., while the  $p \leq 0.05$  was considered statistically significant.

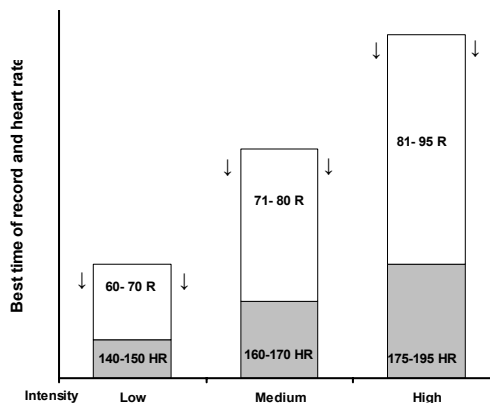


Fig. 1. Time line of experimental design. ↓ indicated saliva sampling=% best swimming record. HR=Heart rate.

**RESULTS**

Mean saliva [C], [DHEA] and [DHEA:C] are presented in Fig 2. For none of the swimmers did the [C] change significantly during the study.

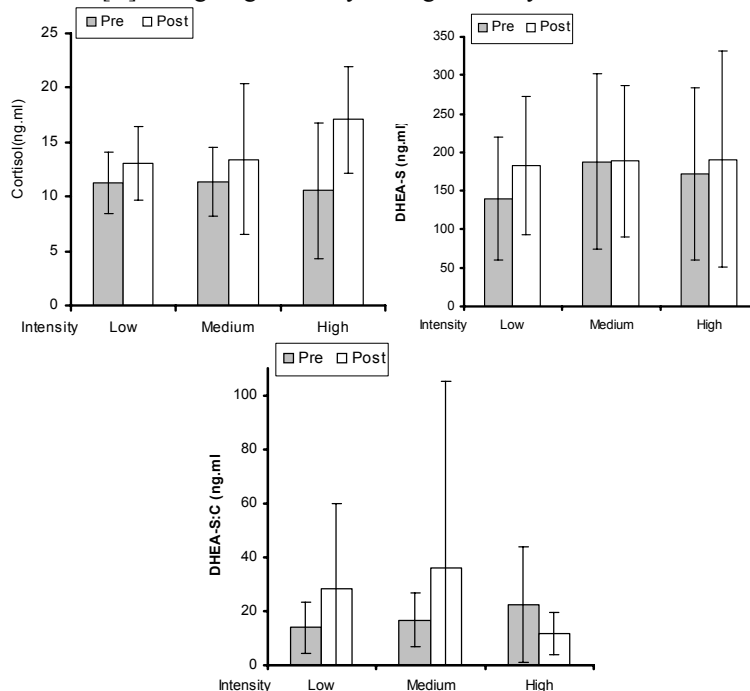


Fig. 2. Average of salivary cortisol, DHEA-S and DHEA-S:C concentration during various stages of sampling. Data are expressed as mean± standard deviation.

An analysis of the results of the comparison of intensity and exercise time and their effect on saliva cortisol concentration shows that exercise intensity ( $F=0.62$ ,  $p=0.54$ ) and exercise time had no significant effect. In addition, the interaction between intensity and exercise time was not significant.

Exercise intensity and exercise time had no significant effect on saliva DHEA concentration. In addition, combinations of intensity and exercise are not significant.

Variation analysis results regarding the effect of intensity and exercise time on saliva relation of (DHEA):(C) show that exercise intensity and exercise time had no significant effect. In addition, the combination of intensity and exercise time is not significant.

## DISCUSSION

The first finding of this research shows that exercise intensity has no significant effect on saliva cortisol concentration.

In this research, saliva cortisol concentration was studied for any increase after low, moderate and high intensity exercise, and its values following exercise were compared to those before the exercise. Nevertheless, the increase was not significant in statistical terms. The greatest increase in cortisol concentration was seen after high intensity exercise and the smallest following moderate intensity exercise. There are various mechanisms that explain the reason of saliva cortisol concentration increase after exercise of different intensity. One of these mechanisms is hormone secretion increase through stimulation of the Hypothalamus. The hypophysial-adrenal axis leads to ACTH secretion increase from the hypophysis, and as we know, increase of ACTH secretion is the most important factor for stimulating cortisol secretion (Guyton & John, 1996).

Filaire et al. compared saliva cortisol concentration between the best female swimmers and the best female handball players, and showed significant increase but there was no significant increase among the swimmers (Filaire et al., 1998). The present research results and the cited research results are the same.

The other mechanism affecting cortisol concentration change is the result of a difference between hemodynamical varieties of the exercise environment. Nilsen et al. reported (1984) that, during swimming, the blood volume from the lower blood vessels moves to the upper parts of the body (Filaire et al., 1998). Guezennec et al. (1986) showed that changes in blood volume due to resting on the back and water pressure during swimming, lead to changes in hormone responses at levels similar to those of body exercises. In this research, the selected participants completed 6 weeks of exercise; therefore, the mentioned reasons may have been responsible for the lack of changes in cortisol concentration.

Mental stress is one of the mechanisms that affect cortisol hormone secretion from the layers of the adrenal cortex. Most researchers believe that water as a large mechanism, has a great effect on stress decrease.

We can say that because mental pressure which was decreased among the participants due to exercising in the water may be the point which is responsible for a lack of change in saliva cortisol concentration. Jacks et al. (Jacks et al., 2002) observed the impact of training at 3 different levels of intensity on salivary cortisol and deduced that the salivary cortisol density during breaks and during training sessions of low and medium intensity did not make a significant difference, while there was a significant difference after the training session with high intensity. The findings of our research were compatible with these in terms

of cortisol density after a high intensity training session. Pantelidis (1997) and O'Connor (1987), studying the effects of short-term sports activities on saliva cortisol concentration, suggested that exercise does not have a significant effect on saliva cortisol concentration (O'Connor & Corrigan, 1987; Pantelidis et al., 1997). In addition, Duclos evaluated corticosterone axis sensitivity in athletes who did endurance exercise after exercising, and found a cortisol increase (Duclos et al., 1998).

The present research findings and those of Filaire, Pantelidis, O'Connor, Nilsen and Guezennes are the same, but differ from those of Duclos. In the majority of these studies, the results state that a change in cortisol concentration is related to intensity, duration, type of exercise environment, fitness level and mental pressure (Edward & Donald, 1999).

In addition, Kiess et al. showed that cortisol concentration is related to the age of the subjects (Kiess et al., 1995). Considering the cited facts, it is possible to claim that the reason for this stated difference is due to the difference in type, environment, intensity, length of the training, level of physical fitness, age and gender of the subjects, their ability to withstand psychological pressure and their diet.

Other findings of the present research showed that exercise intensity does not have a significant effect on saliva DHEA concentration.

In this research, saliva DHEA concentration was increased after low, moderate and high intensity exercise, but this increase was not significant in statistical terms. DHEA is one of the specific adrenal hormones and is controlled by ACTH.

It is not obvious that its increase is due to the very low DHEA metabolic clearing or stimulation of glucocorticoid and androgen secretion of the adrenal cortex (Duclos et al., 1998).

One of the mechanisms that led to a change in DHEA secretion was the secretion control of this hormone by hypothalamus and hypophysis-adrenal axis (Jacks et al., 2002). The DHEA hormone is one of the most important steroid hormones in the human body.

DHEA activity creates an Anabolic effect on several kinds of tissue by transforming into sexual steroids (testosterone and estrogen), as testosterone and DHEA have anabolic effects and are controlled by ACTH and cortisol (Mackinnon et al., 1999). Physiological hypotheses show that DHEA concentration specifies an anabolic condition in endocrinology and maintains it (Chatard et al., 2002). The increase in the response of the hypophysis during high intensity exercise has a positive effect on androgenic-anabolic activity, while physiological pressure decreases with a decrease in exercise. In addition, exercise load, diet and cortisol concentration are important and effective factors in DHEA secretion (Chatard et al., 2002; Filaire et al., 1998; Filaire et al., 1996; Mackinnon et al., 1999).

#### **The relation between the training intensity and the salivary DHEA concentration**

The conclusions drawn in this research have shown that the DHEA density did not change following training sessions at different levels of intensity, which differs from the previously reported changes in testosterone, even though the DHEA is the inner testosterone constructor. Mojica et al. came to the conclusion that the T intensity has a positive relation with the LH following a reduction in training after an intensive swimming training session (Chatard et al., 2002). It has been confirmed that the increase in the hypophysis responses during an intensive training session has a positive effect on the androgenic-anabolic activity during a reduction in the physiological pressure period such as the reduction of training or the reduction of the period of training (Walsh et al., 1998). In addition, the training intensity, the dietary and cortisol density are important

and influential factors in the T secretion (Cumming et al., 1983, Volek et al., 1997). In 1998, Filaire et al. studied cortisol density and DHEA following a sixteen-week program involving women and came to the conclusion that the DHEA density was significant elevated following a training session. The current research results are in contrast with the above results which can be a result of the difference in intensity, type, length and the environment of training and also the condition of the subjects' diet and the change in cortisol density, since these factors have a great effect on the density of DHEA. In another study, Filaire & Lac (2000) studied the level of testosterone gathered and observed in the salivary DHEA among female handball players. The researchers concluded that training did not cause a significant difference in the salivary DHEA and T density (Filaire & Lac, 2000), which shows that our current results are not compatible with the stated ones, which may be due to the similarity of the gender of the subjects who were all female participants, in spite of the difference in training design and environment. The present research findings and the research findings of Chetard, Filaire & Lac (2000) are the same but contrary to the findings of Filaire (1998). The reason for the similarity between the present research findings and Chatard's findings were similarities in terms sex, exercise environment and sport of the subjects and the reason for the similarity between the present research findings and Filaire & Lac's findings was similarity in terms of the sex of the subjects.

The reason why the present findings and Filaire's findings differ is the difference between intensity, duration and type of exercise, exercise environment, diets of the subjects and changes in cortisol concentration, since these factors have an effect on DHEA concentration. The other finding of this research shows that exercise intensity has no significant effect on the saliva (DHEA):(C) relation. One of the mechanisms that result in changes to the (DHEA):(C) relation is the change in the testosterone and cortisol amounts. Mujika and his colleagues (1996) recognized that the T:C relation is inversely proportional to the number of weeks spent swimming (Mujika et al., 1996).

Castil and his colleagues (1991) reported a significant increase in cortisol concentration and a decrease in T concentration, after doubling the exercise distance of swimmers (Dimitriou et al., 2002). Some of the researchers suggested that volume, intensity changes and exercise on land do not have an effect on the T:C relation (Tanaka et al., 1993).

### **The relation between the training intensity and the salivary [DHEA]: [C] RATIO**

Gilian found that hormonal responses in male and female swimmers are different. For instance when the swimming length increases by fifty percent per day, the cortisol density in women elevates and decrease in men, but the T intensity does not change in women (Gilian et al., 1996). There are few studies about the (DHEA):(C) relation in swimmers, but there are some studies regarding the T:C relation which transforms the shape of the (DHEA):(C) relation and may affect a change in the Anabolic-Catabolic balance (Fry & Kraemer 1997; Urhausen et al., 1995). Even though the six-week training program is not long, other physiological features such as technical and metabolic changes, which were not measured in this research, might be influential. In 1993, Snegoraskaya and Viro found that not only can a twenty-year training program cause a progress in performance, but it can also increase the level of the cortisol (Streenberg et al., 1997).

Based on the fact that cortisol concentration affects the (DHEA):(C) relation, and in this research is in contrast with Filaire's research findings, we can say the reason for this

variety is the duration and type of exercise, type of sport, exercise environment (water and land), age, subjects' physical fitness, exercise intensity and changes in the cortisol and DHEA. Our findings are similar to Chetard's findings because the exercise environment and sex of the subjects were identical.

#### CONCLUSION

The present research findings show that a 6-week swimming exercise program does not lead to a significant increase or decrease in the cortisol and saliva DHEA concentration. In addition, a 6-week exercise program does not lead to a disturbance in the Anabolic – Catabolic balance.

Therefore, according to the findings of this research, we suggest that if the exercise program is long, athletes should exercise at low intensity to maintain the anabolic – catabolic balance and reinforce the mucous immune system to improve sport results.

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## **EFEKTI RAZLIČITOG INTENZITETA VEŽBANJA NA KONCENTRACIJU STEROIDA U PLJUVAČKI VRHUNSKIH PLIVAČICA**

**B. Yazdanparast, M.A. Azarbayjani, M.J. Rasaei,  
Morteza Jourkesh, Sergej M. Ostojić**

*Svrha ove studije je bila da se utvrde efekti različitih intenziteta vežbi (nizak, srednji i visok) na koncentraciju Kortisola i DHEA u pljuvački vrhunskih plivačica. 10 mladih plivačica su dobrovoljno izabrane za ispitanice. Ispitanice su učestvovala u šestonedeljnom programu vežbi. Program vežbi je bio podjeljen u tri vremenska dela. Deo osnovne izdržljivosti je izvođen u prve dve nedelje, deo specijalne izdržljivosti u druge dve nedelje, i deo sa teškim vežbama u poslednje dve nedelje. Uzorci pljuvačke su sakupljeni na kraju svake dve nedelje, pre i odmah posle vežbanja studijske grupe. Koncentracija steroida u pljuvački je utvrđena korišćenjem ELISA metoda. Rezultati istraživanja su pokazali da: vežbe sa niskim, srednjim i visokim intenzitetom nemaju značajan efekat na koncentraciju kortisola, DHEA i razmeru DHEA u pljuvački C vrhunskih plivačica  $p > 0.05$ .*

*Nije postojala značajna korelacija između koncentracije sa kortisolom u pljuvački nakon vežbanja sa visokim i niskim intenzitetom  $p > 0.05$ . Ali negativna značajna korelacija je pronađena između koncentracije sa kortisolom u pljuvački nakon vežbanja sa srednjim intenzitetom  $p < 0.05$ . Nije postojala značajna korelacija između koncentracije sa razmerom DHEA u pljuvački: C nakon vežbanja sa niskim, srednjim i visokim intenzitetima  $p > 0.05$ . U zaključku rezultati ove studije pokazuju da steroidi u pljuvački ne utiču na intenzitet vežbanja.*

**Ključne reči:** Kortizol, DHEA, Pljuvačka, Plivač, Trening