

## **Examining certain oxidative stress parameters of coaches of Turkish national boxing team before and after the matches**

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### **ABSTRACT**

*Purpose: This study is performed to determine effects on certain oxidative stress parameters (malondialdehyde, 8-Isoprostane, 4-hidroksinonenal, 8-hidroksi-2-deoksiguanozin) and vitamin C levels of Coaches of Turkish National Boxing Team before and after the matches of International Boxing Championship. 24 male coaches, who train national boxing teams of big male boxers (12) and big female boxers (12) constitute the research group. Blood samples of coaches participating in the international boxing tournament were taken twice being before and after the matches. Data collected before the matches and laboratory results were utilized as covariant. Relative changes in the data obtained after the matches were analyzed in the SAS package program by utilizing PROC MEANS procedure. Significance level of changes occurred is determined by means of Paired t test that adds significance in the level of  $P < 0.05$ . According to the data, the difference in the values of MDA level before and after the match was statistically important ( $P < 0.001$ ). The difference in the pretest and posttest measurements of 8-ISO levels was also important ( $P < 0.05$ ). The difference found between the pretest and posttest measurements of 4-HNE levels was statistically significant ( $P < 0.05$ ). However, the difference in the values of 8-OHdG and Vitamin C parameters before and after the match was not important statistically ( $P > 0.05$ ). It can be stated that boxing matches because important changes in the values of certain oxidative stress parameters being Malondialdehyde, 8-Isoprostane, 4-hydroxynonenal values of national boxing team coaches according to the results obtained.*

**Key words:** Coach, Oxidative Stress, Malondialdehyde, 8-Isoprostane, 4-hydroxynonenal, 8-hydroxy-2'-deoxyguanosine.

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### **INTRODUCTION**

Boxing coaches experience both physical and mental fatigue. It is obvious that coaches are also exerting great efforts for the sporters to be successful since national team camps are the places where boxers are subjected to an intense training program.

Oxygen utilization rate increases 10-20 times systematically during walking and jogging [1] and it increases 100-200 times in the skeletal muscles [2]. This increase in the oxygen utilization rate in the mitochondria known as the energy generating center of the cells is concluded by the triggering of the formation of free radicals and other reactive oxygen species (ROS), and thus by the increase of ROS release from the mitochondria during exercises [3]. Excessively generated ROS causes lipid peroxidation and oxidative stress which led to DNA damage [4]. Therefore, primary source of oxidative stress is known as mitochondria [5]. Levels of malondialdehyde (MDA) and 4-hydroxynonenal (HNE) that are products of lipid peroxidation increase in the human serum depending on the physical activity. Accordingly, decrease is observed in the GSH and antioxidant enzyme levels in line with the increase in these levels. Very fast HNE metabolism reveals the role of HNE degrading means as an important part of secondary antioxidative defense mechanisms in order to prevent modification of proteins by the oxidative stress

products [6]. Lipid peroxidation may be evaluated by the measurement of primary or secondary peroxidation products. Primary products are hydroperoxides and conjugated products. Secondary products are thiobarbituric acid, reactive agents, gas alkenes and F2-isoprostans. For the present, among these indicators, F2-isoprostans are accepted as the most accurate indicator of in vivo oxidative stress in human in line with the current methods [7]. Production of free radicals exceeds the antioxidant capacity in cases necessitating overexert, and consequently the said oxidative stress state is experienced. Superoxide dismutase (SOD) and glutathione peroxidase increase their activity as antioxidant enzymes and provide protection against oxidative stress. Isoprostans, which are indicators of oxidative stress, are members of prostaglandin isomer family and are formed by the oxidative modification of polyunsaturated fatty acids and free radical catalyzed mechanism. Differently from classic prostaglandins which are formed with the effect of cyclooxygenase enzyme and free arachidonic acid (AA), isoprostans may form from arachidonic acid in the esterified form in membrane phospholipids. Formation of lipid peroxidation products such as 4-hydroxynonenal (4-HNE), malondialdehyde (MDA) and 8-hydroxy-2-deoxyguanosine (8-OHdG) causes inhibition of mitochondrial respiratory chain [8, 9]. Stress, not so easy to identify, preferred to be described, is one of the most complex concepts today [10].

When unneeded efforts and excitements of coaches increase their oxidative stress, performance of boxers may be affected adversely. Concentrations of coaches should be at top level especially in the World, Olympiad and European championships. With this aim, certain oxidative stress parameters, being 8-Isoprostane, 4-hydroxynonenal, 8-hydroxy-2-deoxyguanosine, and vitamin C levels of male boxing coaches of male and female national boxing teams are examined by means of blood samples taken before and after the matches of International boxing championship.

### MATERIALS AND METHODS

24 male coaches, who train national boxing teams of big male boxers (12) and big female boxers (12) constitute the research group. Blood samples of coaches participating in international boxing tournament were taken before and after matches from their forearm vein into gel biochemistry tubes by means of 10 cc injectors, and they were centrifuged on Hettich trade mark Universal 320 model after waiting time is completed. After serums of blood samples received are eluted by centrifuge at 4000 rotations for 5 minutes, they were taken into eppendorf tubes and they were preserved in Hettich freezer in the Faculty of Veterinary Science, Department of Animal Nutrition at -80 °C till the time of analysis. MDA levels in the serum samples were analyzed according to the method specified by Karatepe as the indicator of lipid peroxidation [11].

8-ISO (Cat: 516351, Cayman Chemical Company, Michigan, ABD), 4-HNE; Cat: 0903339, Cell Biolabs Inc., San Diego, ABD) and 8-OHdG; Cat: 589320, Cayman Chemical Company, Michigan, ABD) levels in the serum samples were determined by means of ELISA device (Elx-800; Bio-Tek Instruments Inc, Vermont, ABD.) and by using commercial ELISA kits. C Vit. Measurement was made in TERMO HPLC" by utilizing commercial testing kits. Vitamin C peaks were obtained at 254 nm by adjusting 0.75 ml/minute flowing rate at HPLC Bischoff Prontosil AQ 5µm column.

Data collected before the match and laboratory results were utilized as covariant. Relative changes in the data obtained after the match were analyzed by SAS [12] package program with the utilization of PROC MEANS procedure. Significance level of changes occurred was determined with the aid of Paired t test which adds significance in the level of  $P < 0.05$ .

### RESULTS

When we examine oxidative stress parameters of coaches before and after the match MDA is found as  $1.01 \pm 0.39$  µmol/L before match, and as  $0.12 \pm 0.09$  µmol/L after match; 8-ISO is determined as  $79.49 \pm 10.01$  pg/ml before match and as  $95.45 \pm 9.38$  pg/ml after match; 4-HNE is found as  $8.79 \pm 0.16$  nmol/L before match and as  $8.59 \pm 0.23$  nmol/L after match. There are statistically significant differences between the measurements. 8-OHdG is determined as  $1.84 \pm 0.30$  ng/ml before match, and as  $1.84 \pm 0.28$  ng/ml. after match; C Vit is found as  $1.20 \pm 0.54$  mg/ml before match and as  $0.93 \pm 0.32$  mg/ml after match. No statistically significant difference is found in these parameters (Table 1).

Table 1: Blood and oxidative stress levels of Coaches (n=24)

Parameter	Before match	After match	Difference	t value	P
	Mean $\pm$ SD	Mean $\pm$ SD	Mean $\pm$ SD		
MDA ( $\mu$ mol/L)	1.01 $\pm$ 0.39	0.12 $\pm$ 0.09	0.90 $\pm$ 0.30	9.09	0.0001**
8-ISO (pg/ml)	79.49 $\pm$ 10.01	95.45 $\pm$ 9.38	-15.96 $\pm$ 9.73	-0.93	0.0349 *
4-HNE (nmol/L)	8.79 $\pm$ 0.16	8.59 $\pm$ 0.23	0.19 $\pm$ 0.19	2.14	0.0490 *
8-OHdG (ng/ml)	1.84 $\pm$ 0.30	1.84 $\pm$ 0.28	0.01 $\pm$ 0.29	0.04	0.9672 -
C Vit (mg/ml)	1.20 $\pm$ 0.54	0.93 $\pm$ 0.32	0.27 $\pm$ 0.46	1.71	0.0991 -

- :  $P > 0.05$  \* :  $P < 0.05$  \*\* :  $P < 0.001$ 

(MDA)malondialdehit, (8-ISO) 8-isoprostane, (8-OHdG) 8-hydroxy-2'-deoxyguanosine ve (4-HNE) 4-hidroksinonenal

## DISCUSSION AND CONCLUSION

In the research certain oxidative stress parameters of coaches of Turkish national boxing team measured before and after the matches; and MDA average of the research groups is specified as 1.01 $\pm$ 0.39  $\mu$ mol/L before match, and as 0.12 $\pm$ 0.09  $\mu$ mol/L after match; and a decrease is observed between the values measured before and after the match. This decrease is found to be statistically significant (Table 1). When similar studies are examined [13, 14, 15, 16]; a significant decrease is observed in the MDA values after exercising in comparison to the values before exercising. In another study, Pala et al [17], have determined decrease in the MDA values of boxers before and after the match. Şinforoğlu [18] has stated that decrease in MDA values of sporters before and after the aerobic sportive loading made before the preparation stage is not statistically significant. Different findings can be discovered in the literature, it is determined that MDA did not change in acute exercising [19], no significant effect is observed in MDA values of young trained Judaists [20], MDA did not change in aerobic exercising [21], no change is observed in MDA in maximal exercising [22].

Average of 8-ISO, as a reliable indicator of oxidative stress, is determined as 79.49 $\pm$ 10.01 pg/ml before match, and as 95.45 $\pm$ 9.38 pg/ml after match; and increase observed between measurements before and after match is found statistically significant (Table). When similar studies are examined, it is stated that 8-ISO parameter has increased in a studies performed on healthy young people and healthy Japanese individuals [23, 24]. In another research, decrease seen in the 8-ISO parameter before and after camp of boxers of Turkish national boxing team is found to be statistically significant [17]. Information in the literature supports our study.

4- HNE average as another indicator parameter in the study is determined as 8, 79 $\pm$ 0.16 nmol/L before match and as 8.59 $\pm$ 0.23 nmol/L after match; and decrease between these values is found statistically significant (Table). In similar studies, a significant decrease is determined in the 4-HNE values of boxers before and after camp [17], and even though 4-HNE values of weight lifters decreased after exercising, this decrease did not constitute a statistically significant difference [25].

8-OHdG average is determined as 1.84 $\pm$ 0.30 ng/ml before match and as 1.84 $\pm$ 0.28 ng/ml after match, and the difference is not statistically significant (Table). In the literature; it is stated that decrease is observed in the values of boxers before and after the camp, however this difference is not statistically important [17]. This finding shows parallelism with our study. In other researches, 8-OHdG value of sporters doing wrestling exercises is found to be lower and significant in comparison to the sedentary individuals [26]. Asami et al., have determined in their study where sporters and untrained individuals are compared that values of sporters are lower [27]. This literature information differs from our study.

C Vit is determined as 1.20 $\pm$ 0.54 mg/ml before match and as 0.93 $\pm$ 0.32 mg/ml after match. There is no statistically important difference in this parameter (Table). The hypothesis in this study is to find whether or not stress observed in coaches before and after match causes any oxidative damage. Literature review realized on this subject supports the significant differences in the oxidative stress parameters of coaches in the matches.

It can be said that boxing matches because important changes in the values of certain oxidative stress parameters being Malondialdehyde, 8-Isoprostane, 4-hydroxynonenal values of national boxing team coaches according to the results obtained.

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