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THE EFFECT OF SIX WEEKS SPRINT TRAINING ON SERUM ANTIOXIDANT LEVELS IN SOCCER PLAYERS

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Abstract:

The aim of this study was to determine the effect of six week sprint training applied to soccer players on antioxidant levels. Eighteen football players participated voluntarily and six-weekly sprint training was applied. Soccer players were given a 20-m sprint test to determine antioxidant levels before training, and blood samples were taken before and after the test and this were repeated after six weeks. The effects of the training to the antioxidant systems were searched by the estimated the levels of SOD, CAT and MDA on the taken blood samples before and after the sprint training. The statistically significant results were obtained for serum CAT (2,89±0,85 and 8,42±0,81 U/ml) and MDA $(16,39\pm2,66 \text{ and } 29,10\pm2,62 \text{ mmol/L})$ (p<0,05) values before and after the sprint test of the athletes at the end of the 6-week sprint training, but there was no statistically significant result despite the difference in SOD (1,74±0,13 and 2,49±0,13 U/ml) value. On the other hand statistically significant results were found in serum SOD (1.67±0.36 and 0,88±0,20 U/ml) and CAT (0.15±0.01and 5,25±0,47 U/ml) (p<0,05) values before and after sprint test of athletes, but there was no statistically significant result even though there was a difference in MDA (4.83±0.99 and 3,68±0,77 mmol/L) value. Consequently, making training can cause development on antioxidant defense, and it can affect the antioxidant production.

Keywords: sprint exercise, superoxide dismutase, catalase, malondialdehyde levels, soccer players

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1. Introduction

Physical activities such as walking running, jumping can be possible for organism by means of the spasm of the muscles of skeleton. Aerobic organism is often face to face free radicals that their toxins are high even in normal and peaceful life free oxygen derivatives are often produced in low levels during the anaerobic metabolism. The amount of free oxygen derivatives that forms with the increase of metabolism activity increases (Akdemir et al. 2016; Maughan et al 1989). Cells and the organism as a whole have the antioxidant systems against the destructive effect of free radicals. These mechanisms influence by putting the premise materials of free oxygen radicals or cleaning the free radicals. ROS are continuously produced during normal physiological events and can easily initiate the peroxidation of membrane lipids, leading to the accumulation of lipid peroxides (Gülçin, 2010; Gülçin and Daştan, 2007; Balaydın et al., 2010).

During the physical exercise, the speed of metabolism raises proportional to the intensity of muscle activity. The level of oxidative damage which can occur during physical training is not only determined by the formation of free radicals, but also by the defence capacity of antioxidants. While it is expressed that especially acute, intensive training may cause oxidative stress, it is suggested that regular endurance training can reduce oxidative stress and muscle damage after the exercise, and develop antioxidant defence capacity. (Tas, 2011)

Exercise can cause oxidative stress according to its intensity and period. Thus, it is thought that lipid pre-oxidation confirms if the increase in the level of the free oxygen radicals passes the antioxidants in the capacity of the defence of cells during the exercise. (Leaf, 1997; Akdemir et al. 2016) MDA that is one of the materials appears as a result of lipid peroxidation is used as an indicator of oxidative stress. It can be thought that the amount of damage that is formed on body can affect the regeneration period. The damage of oxidative that can be formed during the physical exercises can be determined not only the production of free radicals but also the defence capacity of antioxidant superoxide dismutase (SOD), katalase (CAT) and malonaldehyde (MDA) provide the first defence line against the reactive oxygen species (ROS) produced during the exercise. Therefore, it is thought that exercises will affect these enzymes directly.

In this study was planned to determine the effect of six week sprint training applied to soccer players on antioxidant levels.

2. Materials and methods

2.1 Participants

This study was made on 18 amateur soccer players who are healthy and their age is between 19-30 years and active in soccer. In this study the aim of the study with the object and possible risks are explained and their written approval were taken.

2.2 Training program

The 6-week sprint interval training program used for all subjects is presented in Table 1. All distances sprinted (at either 90% or 100% of maximum speed) were between 30 and 80 m, with 90% of the training performed over 40±60 m. In the final 3 weeks of training all sprints were over 60 m or less. Progressive overload was applied by increasing the number of sprint repetitions in a session from an initial 22 to a high of 42 in two of the sessions in the 6th week, and by performing more sprint sets at 100% of maximum speed. A gradual reduction in the work:recovery ratio was also applied from an initial 1:6 down to 1:4 on certain sets. The recovery time between sets of sprints was 2±4 min with the longer recovery time allowed between sets of maximum effort. Slow walking and jogging was performed in the recovery periods between sets. When sprint sets at 90% of maximum speed were performed (see Table 1) the experimenter kept time on each sprint as a check on the required intensity. All training sessions were supervised by the same experimenter and were completed in small groups of three or four, except for two occasions when one subject trained without supervision.

Table 1: The sprint training program, showing the number of repetitions and distances covered for each session. Five subjects completed 16 sessions while the other 4 subjects each completed 14, 15, 17 and 18 sessions, respectively. Each subject completed at least three sessions of 40±42 repetitions. Where a set of intervals are underlined those efforts were run at maximum speed. (W:R) (work:recovery ratio) (Dawson et al. 1998).

Week	Session							% Maximum effort	W:R	Number of reps.
1	1	6 × 80	6 × 60	6 × 40	4 × 40			90	1:6	22
	2	6×80	6×60	6×40	4×40			90	1:6	22
	3	6×80	6×60	6×40	6×40			90	1:6	24
2	4	6×80	6×60	6×40	8×30			90	1:5	26
	5	6×80	6×60	6×40	8×30			90	1:5-6	26
	6	4×80	6×50	8×40	6×40	6×30		90/100	1:5-6	30
3	7	4×80	6×50	8×40	6×40	6×30		90/ <u>100</u>	1:6	30
	8	8×30	6×50	8×30	6×40	6×30		90/100	1:5-6	34
	9	8×30	6×50	8×30	6×40	6×30		90/ <u>100</u>	1:5-6	34
4	10	$\frac{8 \times 30}{6 \times 60}$	$\frac{8 \times 50}{8 \times 50}$	6×40	8×50	6×60		$90/\overline{100}$	1:5	34
	11	6×60	8×50	6×40	8×50	6×60		90/ <u>100</u>	1:6	34
	12a	6×60	8×50	6×40	6×40			90/100	1:5	24
5	13	8×50	8×40		8×40	8×50		$90/\overline{100}$	1:5-6	40
	14	8×50	8×40	$\frac{8 \times 40}{8 \times 30}$	8×40	$\frac{8 \times 50}{8 \times 50}$ 8×50		90/100	1:5-6	40
	15	8×50	8×40	8×30	8×40	8×50		90/100	1:4-6	40
6	16	8×50	8×40	8×30	6×50	6×40	6×30	$90/\overline{100}$	1:4-6	42
	17	8×30	8×40	8×50	6×50	6×40	6×30	$90/\overline{100}$	1:4-6	42
	18	8 × 30	8×40	6×50	6×50	6×40	6×30	90/100	1:4-6	40

^a:Very windy day-session reduced

2.3 Sprint tests

The participants performed three maximal 30m sprints (with 5m and 20m split times also recorded). During the 3min recovery periods in-between, the participants walked back to the starting line and then passively waited for the next sprint. Time was recorded using three photo-cell gates (Brower Timing Systems, Salt-Lake City, UT, USA, accuracy of 0.01 s) positioned 5, 20 and 30m from the starting line at a height of 1 m. The participants commenced the sprint when ready from a standing start 0.5m behind the first timing gate. Stance for the start was consistent for all participants. The

best (fastest) 30m sprint time and the associated 20m sprint times were selected for analysis.

2.4 Measure of Height and Weight

On the smooth floor, the height measures of test group sportsmen were taken as cm using the tape measure and their weight was taken as kg by sensitive bascule.

Table 1: Age length and weight of the soccer players

Variables	N	Minimum	Maximum	Average
Age	18	19.00	30.00	23.53 ± 3.6
Length	18	1.60	1.83	1.73 ± 0.05
Weight	18	60.00	75.00	67.84 ± 5.1

2.5 Blood Analysis

Blood samples were taken twice just before and after the exercise and these blood samples were taken again twice after training period of 6 weeks. The levels of serum SOD, CAT and MDA were determined on the neparinize blood samples taken from venous. The blood samples were taken before and after burden.

2.6 Taking Blood Samples

Blood was collected from the antecubital vein at the fasted state at 07:30–08:00 a.m., approximately 20 ml to each of 2 sample tubes. Some donations were used to determine the parameters of blood count with differential. The remaining plasma samples were collected, frozen at -80 C, and kept in these conditions until biochemical measurements. Determination of the cellular components of blood was conducted on a hematology analyzer BC-2800 Mindray's. Blood smears were evaluated independently. The activity of catalase (CAT) was determined by the set of reagents OxiSelect Catalase Activity Assay Kit, (No. Cat. STA-341) produced by the Cell Biolabs company. The activity of superoxide dismutase (SOD) and malondialdehyde concentration was performed using reagents from kits Cayman Chemical Company: Superoxide Dismutase Assay Kit (No. Cat. 706,002), Antioxidant Assay Kit (No. Cat. 709,001) and TBARS Assay kit (No. Cat. 10,009,055). All assays were performed strictly according to the instructions included by the manufacturer.

2.7 Statistical Analysis

Descriptive statistic (average and SD) belong to the soccer player in research field were made test was applied for the comparison of biochemical and 20m sprint test values taken before and after sprint exercise SPPS 21 statistic program was used for the analysis of research data.

3. Results

Table 2: Serum SOD, CAT and MDA levels before and after sprint testing of athletes prior to Sprint training

Variable	N	SOD (U/ml)	CAT (U/ml)	MDA (mmol/L)	p
Before 1		1,67±0,36*	0,15±0,01*	4,89±0,99	m<0.0E
After 1	18	0,88±0,20*	5,25±0,47*	3,68±0,77	p<0,05

^{*:} p<0,05

When Table 2 was examined, statistically significant results were found in serum SOD $(1.67\pm0.36 \text{ and } 0.88\pm0.20 \text{ U/ml})$ and CAT $(0.15\pm0.01 \text{and } 5.25\pm0.47 \text{ U/ml})$ (p<0.05) values before and after sprint test of athletes, but there was no statistically significant result even though there was a difference in MDA $(4.83\pm0.99 \text{ and } 3.68\pm0.77 \text{ mmol/L})$ value.

Table 3: Serum SOD, CAT and MDA levels before and after sprint testing of athletes after Sprint training

Variable	N	SOD (U/ml)	CAT (U/ml)	MDA (mmol/L)	p
Before 2		1,74±0,13	2,89±0,85*	16,39±2,66*	<0.0F
After 2	18	2,49±0,13	8,42±0,81*	29,10±2,62*	p<0,05

^{*:} p<0,05

When Table 3 was examined, statistically significant results were obtained for serum CAT ($2,89\pm0,85$ and $8,42\pm0,81$ U/ml) and MDA ($16,39\pm2,66$ and $29,10\pm2,62$ mmol/L) (p<0,05) values before and after the sprint test of the athletes at the end of the 6-week sprint training, but there was no statistically significant result despite the difference in SOD ($1,74\pm0,13$ and $2,49\pm0,13$ U/ml) value.

Table 4: 20-m sprint values of soccer players before and after Sprint training

Variable	N	Ortalama	t	P
Before	_	3.10 ± 0.11	1 0/1	D<0.05
After	18	2.95 ±0.19*	-1.061	P<0.05

^{*:} p<0,05

Before the Sprint trainees, the 20-m sprint values of footballers were 3,10±0,11 seconds on average and 2,95±0,19 seconds after sprint training. It was determined that the result obtained was statistically significant (p<0,05).

4. Discussion

They are very important to protect the present balance between the oxidant and antioxidant systems and the structural totality of cells and fabrics and to make their normal functions. Antioxidant system is insufficient against the oxidative stress that some disease in organ and fabrics or other factors (as exercise) increase too much and this makes illness and also causes that several complication appears fast.

Antioxidant enzymes, nutritious for body, use too effective antioxidant defence system such as CAT; SOD or MDA to reduce the oxidative stress and to impute free radicals. If free radical levels that passed the antioxidant capacity, fats, proteins and other cell parts are oxidized (Clarkson et al. 2000; Akdemir et al. 2016). Endurance exercise was used in the most of experimental study that oxidative stress, the exercise causes; and its plasma antioxidants or its definite effect on lipid peroxidation was searched.

Heavy physical exercises, causes fast oxygen increases both on whole body and especially on skeleton muscles. Most of the exhausted oxygen is used for mitochondrial and production of ATP. Some studies showed there was a link between the increase of oxygen consumption in physical exercise and production of free radical. For each molecule, reduced respiration, one free radical is supposed to be produced. During the exercise oxygen consumption can increase 10-15 times and, the oxygen amount in the active muscle can increase about 100 multiple (Sen, 1999).

The aim in this study is to make observations about the changes in antioxidant defence after the marked by repetition sprint exercise parallel to energy of metabolism that soccer players use during the match and training; and compare the results from other exercises.

SOD Works in the cell as one of the most important enzymatic antioxidant that struggled against the super oxit radicals (Powers et al. 1999). The increase in SOD enzyme increases the defence against the oxidative stress (Fielding et al. 1997). It is observed that total muscle and SOD activities in mitochondrial for volleyball players who made training is higher than others who did not make training. Marzatico et al. (1997) saw an important increase in higher amount of erythrocytes SOD activity, in his study about short distance runners and marathon runners. An important increase in SOD activity was seen when the short distance runners finish a running exercise and marathon runners run as half marathon level. With this, each study does not give the same result. For example, SOD activity was not seen on athletes who complete the middle intensity bicycle exercise for 8 weeks (Tiidus et al. 1996). In Tauler's study, any change was not seen in SOD activity after the middle intensity of biathlon activity. An increase was seen at the level of erythrocytes SOD for cross-country skiers after the exercise (Hubner-Wozniak et al. 1994). Supramaximal exercise (wingate protocol) that causes fatigue and Zergeroglu et al applied to the sedentary people causes an important increase in the erythrocytes SOD activity.

The obtained SOD values were higher before the sprint test but lower at the end of the test. It may be thought that the level of antioxidant, which is increased by the

level of free radicals during exercise, is inadequate. However, 6 weeks of sprint training increases the level of enzymatic antioxidants, which can be said to have a positive effect on performance.

The fraction of hydrogen peroxide ensuring water and oxygen was done by means of CAT. This antioxidant spans in the cell and mitochondria and peroxisumes occurred in the most of the activity (Maria et al. 2003). Tiidus et al. (1996) reported that the activity of CAT did not change during the 8 weekly aerobic training, they made. Rokitzki et al. (1994) did not determine any change at the level of erythrocytes CAT after the marathon. Aguilo et al. (2000) observed that there was decrease about %20 in erythrocytes CAT on the cyclists in form during the period of 90 minutes exercise. Marzatico et al. (1997) did not observe any change in erythrocytes CAT on runners who made sprint exercise, but they observed on increases in the activity of CAT for long distance runners after 24-28 hours of endurance exercise. Maria et al.(2003) determined that the value of CAT for cyclists decreased. Marzatico et al. (1997) confirmed in their study that little increase was in CAT for sprinters than marathon runners.

In our study, an important increase at CAT level noticed during the speed test made before and after the sprint training. This result is similar to data that the increase of CAT activity will be stabilized with new young erythrocytes, joined the circulation and it can be accepted as an indicator of the exercise adaptation.

It is thought that MDA causes oxidative stress and increases the reactions of lipid peroxidation proportionally with the intensity and period of the exercise (Goldfarb et al. 1994) when the usage of oxygen is low, superoxide radical and its derivatives are influenced with antioxidant defence system. However, in the situation of the exercise that oxygen consumption increases an important level these defence metabolism cannot humour the formation of free radical (Cheeseman et al. 1993). Paradoxical results about MDA have been reported. It has been reported that some part of these differences, stabilized, in MDA activities are bound the change that exercise cause in the plasma volume. Marzatico et al. (1997) found in their studies that plasma MDA on marathon runners who trained at high level and sprinters that trained sprint exercise at high level are high. Kanter et al. (1985) reported that an increase in MDA plasma after the heavy endurance training that they apply to elite sportsmen. Alessio et al. (2000) mentioned that MDA did not change in exhausting aerobic exercise and Duffaux et al. (1997) told that MDA did not show clear increase after the heavy running test, applied to the students of gymnastics and Leaf et al. (1997) reported there were not any change in MDA during the maximal exercise and before and after the exercise in his study. Grisham, (1992) did not found expressive difference in MDA during acute exercise. Dernbach et al. (1993) reported that any change at the level of plasma MDA was not noticed during resting before and after intensity pulling training for 4 weeks. Hubner-Wozniak et al. (1994) found little increase at erythrocytes MDA before and after the training program that do not train 12 male subjects were trained.

Hubner-Wozniak et al. (1994) practised 12 do not train male subject exercise of weekly intensity bicycle ergonometer; and they found a little increase in erythrocytes MDA before and after training. Rokitzki et al. (1994) reported that increase in MDA was

noticed was noticed for runners and skiers who were in form—right after the heavy training. Sahlin et al. (1991) determined the MDA increased during the acute exercise. Lovlin et al. it increased during exhausting exercise, according to Maria et al. (2003) it increased for cyclists.

In our study we made reduction in MDA occurred after the sprint test before sprint training but this situation do not mean as statistically. Besides an important increase at MDA level was noticed on the test, made after 6 weeks.

The reason of this situation is that MDA reflects the degree of lipid peroxidation occurred after the reaction with free radicals and it was reported that the maximal exercise caused forming of an important amount of free radicals (Haliwell et al. 1992, Kanter et al.1985).

5. Conclusion

The studies about this subject do not cohere. Too many factors effect human performance. This situation can be emanated from training level, different kind of exercise and different measure processes made. It has been thought from the data that supramaximal exercise causes the formation of free radical and oxidative stress.

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