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Plasma antioxidant responses and oxidative stress following a 20 meter shuttle run test in female volleyball players

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Abstract

The effect of physical exercise on oxidant stress and antioxidants has been investigated extensively in the last twenty years. Cells continuously produce free radicals under normal conditions during mitochondrial electron transport chain (ETC). Experimental studies have shown elevated metabolic rate by strenuous physical exercise induces oxidative stress and production of excessive amounts of free radicals. Lipid peroxidation occurs when free radicals react with cellular components involving polyunsaturated fatty acid residues of phospholipids which are very sensitive to oxidation. This study aimed to determine plasma antioxidant responses and oxidative stress following a 20 meter shuttle run test in female volleyball players. Ten female volleyball players from the same team, and ten sedentary female ages between 18-24 years old volunteered to participate in this study. They were in good health and 48 hours before the test did not receive any drug or alcohol. None of them had any endocrine, orthopedic problems. Before the study, Informed, written consent was obtained from all the participants after full explanation of the procedures involved. All procedures were approved by the Selçuk University Meram Medical School of Ethical Committee.

20 meter shuttle run test was designed to estimate the maximal aerobic power of athletes performing in sports with frequent stops and starts (eg. Basketball, volleyball, fencing and so on). Findings of our study demonstrate that in both female groups 20 meter shuttle run test leads to production of more reactive oxygen species than the antioxidant systems can scavenge. Decrease in the activities of these antioxidant enzymes may be due to their inactivation caused by the higher production of the free radicals. it seems that the vulnerability of the body to oxidative stress is significantly enhanced after strenuous exercise test.

Keywords: Antioxidant responses, oxidative stress, volleyball players

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Introduction

The effect of physical exercise on oxidant stress and antioxidants has been investigated extensively in the last twenty years. Cells continuously produce free radicals under normal conditions during mitochondrial electron transport chain (ETC). Experimental studies have shown elevated metabolic rate by strenuous physical exercise induces oxidative stress and production of excessive amounts of free radicals. Lipid peroxidation occurs when free radicals react with cellular components involving polyunsaturated fatty acid residues of phospholipids which are very sensitive to oxidation. Malondialdehyde (MDA) is an end-product of lipid peroxidation which is used as marker of oxidative stress. Antioxidant defence system consisting of endogenous enzymes such as catalase (CAT), Glutathione (GSH) and superoxide dismutase (SOD). These antioxidant enzymes ameliorate the harmful effects of the free radicals.

Free Radicals and Oxidative Stress

Atoms and molecules, which have one or more unmatched electrons on atomic or molecular orbital, are referred as free radicals and materials, which prevent free radicals and their reactions as well as which have enzymatic or non-enzymatic structures. Unmatched electron or electrons are determining the reactivity degree of free radicals. The most important thing for aerobic organisms is free radicals or more generally reactive oxygen species (ROS). Mitochondria has an important role on the formation of free radicals. (Dündar and Aslan 1999, Valko et al. 2007).

Antioxidant defence system consisting of such as Superoxide dismutase (SOD), Glutathione (GSH), Glutathione Peroxidase (GPx). These antioxidant enzymes ameliorate the harmful effects of the free radicals. (Limón-Pacheco and Gonsebatt 2009).

Acute Exercise and Oxidative Stress

ROS is formed as a result of increased oxygen consumption on muscles during physical activities (Özçelik and Karataş 2008). Oxygen input of active muscles increases tens of times along with exercise. Main source of the free radical production is mitochondria. Approximately 2-5 % of the oxygen in mitochondria turns into reactive oxygen species during exercise. Reactions, catalyzed by xantine oxidase during exercise, prostanoids,

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neutrophil activation, hypoxia on muscles, reoxygeneration, calcium homeostasis, decayed in the muscles, and ROS is formed due to proteins, including iron. During high intensity exercises (maximum oxygen consumption is more than 60 %) tissue damage and local inflammations occur. Especially after the exercises, during which muscles elongate, neutrophillic activation increases.

After an exhaustive exercise local acidosis is occured. Acidic environment maintains oxygen separation from hemoglobin. Further, in connection with acidosis, iron is separated from transferrine. Hyperthermia due to exercise cause oxidative damages. Malondialdehyde (MDA), carbonyl compounds and LDL (low density lipoprotein) increase is seen on the serum as a result of lipid peroxidation. Formed superoxide radical reacts with nitric oxide to form peroxynitrite. Along with the exercise cortisol and catecholamine levels are increasing in the plasma. In connection with the exhaustive exercises catecholamines in the plasma are subjected to auto-oxidation. After high intensity dynamic exercises, in connection with the endothelial hyposis, cellular ATP pool exhausts, and ATP dependent calcium ion pump collapses and calcium concentration, which increases within the cell, activates calcium dependent proteases. Activated proteases are increasing the formation of superoxide and hydrogen peroxide.

Blood flow to working muscles during exercise might increase to 40 times more than original. On the other hand, oxygen amount to interior organs, such as liver and kidney, decreases. Formation of free radicals increase as a result of reperfusion of these organs after exercises (Banerjee et al. 2003, 2001, Deaton et al. 2003, Sacheck and Blumberg 2001, Lamprecht et al. 2004).

Acute Exercise and Antioxidant Defense

Increase is seen on the activities of antioxidant enzymes, such as SOD and catalase on muscles, liver and heart tissues after acute exercise (Banerjee et al. 2003).

SOD activity increases on skeleton muscles, heart, liver and erythrocytes during acute exercises. After acute exhaustive treadmill exercise, CuZnSOD and MnSOD enzyme contents and activities are found increased for a period of 1 – 3 days. This finding shows that exercise has a stimulating effect on SOD gene expression (Dinçer 2001). Antioxidant levels differ according to the intensity, type and length of the exercises. During a study made on rats, total antioxidant capacity levels in the plasma reduced after acute exercises. This

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decrease is an evident for huge amounts of increase for free radical formation due to exercise and for insufficiency of antioxidant system in order to cope with free radical production (Fıçıcılar et al. 2003).

Researchers determined that total antioxidant capacity and vitamin C levels decrease in plasma at the end of treadmill effort test, performed on healthy persons who are not performing regular workouts. This decrease on antioxidant capacity shows that oxidant / antioxidant balance shifts in favor of oxidative stress as a result of increased oxidants (Demirbağ et al. 2006).

Regular Exercises Oxidative Stress and Antioxidant Defense

It is observed on perfomed studies that acute aerobic exercise increases oxidative stress production and endogenous antioxidant production increases during chronic aerobic exercise and oxidative stress reduces accordingly (Alessio et al. 2000).

Chronic exercise increases antioxidant defense. It is determined on performed studies that erythrocyte GSH, catalase and glutation reductase activities increase after chronic exercise. (Clarkson and Thompson 2000).

Along with the exercise, it is determined that there is also an increase on antioxidant enzymes. Regularly performed exercise might cause an important adaptation against the oxidative stress. On the bodies of the sportsmen, who implement regular workouts, decrease on formation of reactive oxygen is determined (Brites et al. 1999). This situation can be explained as well adaptation of antioxidant defense systems of sportsmen. It is determined that plasma ascorbic acid, alpha-tocopherol, uric acid and total GSH levels of elite female soccer players, who implement regular workouts, increase just after a soccer game of 40 minutes. This increase on antioxidant defense can be explained with transmission of endogenous antioxidants and / or tissue accumulated antioxidants to circulation along with exercises (Andersson et al. 2009).

Regularly implemented exercises and workouts reduce the harmful effects of free radicals to body and stimulate biosynthesis of antioxidant enzymes (Vina et al. 2000). hile CAT, GPX and Mn-SOD levels of regularly exercised rats on soleus muscle are found higher than non-exercising rats, no difference is determined on their Cu, Zn-SOD levels (Lambertucci et al. 2006).

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Kılıç et al. (2006) had studied affects of exhaustive exercise on thyroid hormones and testosterone levels of elite athletes. At the end of the exercise, while a certain inhibition is determined on the levels of serum thyroid hormones and testosterones, it is seen that 4 week zinc loading eliminates this inhibition (Kılıç et al. 2006).

Methods

Subjects

Ten female volleyball players from the same team, and ten sedentary female ages between 18-24 years old volunteered to participate in this study. They were in good health and 48 hours before the test did not receive any drug or alcohol. None of them had any endocrine, orthopedic problems. Before the study, Informed, written consent was obtained from all the participants after full explanation of the procedures involved. All procedures were approved by the Selçuk University Meram Medical School of Ethical Committee.

Statistical analysis

All data were analyzed using SPSS 13 Statistical Software. Paired Samples-t test was performed. Statistical difference was set at $p \leq 0.05$ (9).

Physical characteristics of the subjects are reported in Table 1.

	Sedentary Group n= 10	**Volleybal Players n= 10**	**t**	**p***
Age	19±1.78	20±1.18	0.734	0.631
Height (cm)	167±2.38	1.81±4.67	2.931	0.084*
Weight (kg)	66±6.14	67±5.73	0.974	0.749
Body Mass Index [(kg/cm^2)]	22±3.2	20±4.6	2.697	0.081*

significant at p<0.05

Study Protocol

On the study day, subjects were driven to Laboratory at the Department of Physical Education and Sports following a 12 hours overnight fast. The 20 meter shuttle run test were performed by the subjects to estimate maximal oxygen uptake ($VO_{2\ max}$).

This test was performed on a hard synthetic surface located in an indoor sports arena. Subjects ran between two lines 20 meter apart in time with a sound signal which was emitted

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from an audio cassette. The frequency of the sound signals increased every minute. The test was terminated when the subject was no longer able to follow the set pace and did not reach the targeted line on three consecutive occasions. The level attained and the number of shuttles at that level allowed a prediction of VO_{2max}, to be made according to Ramsbottom et al (10).

Blood Sampling and Blood Analysis

Blood samples were collected from the antecubital vein of forearm before, Immediately after exercise. Blood samples were collected by vacutainer and immediately centrifuged (3000 rpm for 10 min.) plasma was then obtained and frozen at -80^{O}C for until analysis. Assay kits for determination of MDA, GSH and CAT in plasma samples were used to measure enyzmes activities by spectrophotometry.

Maximal Aerobic Power

Maximal aerobic power ($VO_{2\ max}$), is the highest oxygen (O_2) consumption of the person, reached during sea level performed dynamic exercise under normal conditions, and it is used during determination of aerobic capacity. Increase of O_2 amount, used during unit time, is an indicator for high aerobic capacity, and it is a very important factor, affecting performance during endurance sports. Most reliable test for determination of maximal aerobic power is maximal VO_2 test. This test is effective on determination of cardiorespiratory compatibility. Age, sex, heart flow, arteriovenous oxygen difference (a-v O_2), blood volume, total hemoglobin (Hb) amount and endurance exercises are the effecting factors of maximal aerobic power (Kara and Gökbel 1997, Sınırkavak et al. 2004).

MDA Level Determination

MDA analysis are determined via ELISA Colorimetric method with using Cayman brand (catalogue no: 705002) commercial kits. After mixing with vortex, mouth of the tube is closed and left to 90 ^{o}C water bath for a period of 15 minutes. Tubes, taken from water bath, after leaving into ice for a period of 15 minutes, it is maintained to reach to room temperature. Supernatant is obtained with centrifuging at 3000 rpm for 10 minutes. 2 ml supernatant is put into another tube 1 ml of % 0,675 Thiobarbituric acid (TBA) is added onto it, and left to 90 ^{o}C water bath for a period of 15 minutes. After leaving samples into ice full

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container for a period of 15 minutes, after reaching to room temperature, their absorbance (nmol/ ml) are read at 532 nm on spectrophotometer.

GSH Level Determination

GSH analysis are determined via ELISA Colorimetric method with using Cayman brand (catalogue no: 7003002) commercial kits. All non-sulphidrile groups of erythrocytes are in form of reduced GSH. 5,5'-dithiobis(2-nitrobenzoic acid) (DTNB) has disulphide chromogen structure and reduced by sulphidrile compounds, and forms yellow compound. Absorbance of this reduced chromogene at 412 nm is directly proportional to GSH concentration. 0,2 ml is taken from whole blood and put into 10 m test tube, and 1,8 ml distilled water is added onto it and thoroughly mixed for haemolysis. 3 ml of precipitation solution is rapidly added and mixed. After leaving to room temperature for 5 minutes, it is filtered via raw degree filter paper. After preparation of the tubes, it is closed with the lid, and inverted for 3 times and measured at 412 nm on spectrophotometer within 4 minutes (µ mol/ ml).

CAT Level determination

For CAT level determination Cayman brand (catalogue no: 707002) commercial kit is used. 20 µl hydrogen peroxide is added to the wells, to be studied, and after incubation at room temperature for 20 minutes, 30µl potassium hydroxide and 30µl chromogen are added. After incubation at room temperature for 10 minutes, 10 µl potassium periodate is added and after waiting for 5 minutes, absorbance of each sample are read at 540 nm in wells (nmol / min / ml).

Data Analysis

During calculation of data, SPSS 13.0 statistical package software is used. During in-group assessment, Paired Samples-t test, and during inter group assessment Student t test are applied (Özdemir 2005).

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Findings and Discussion

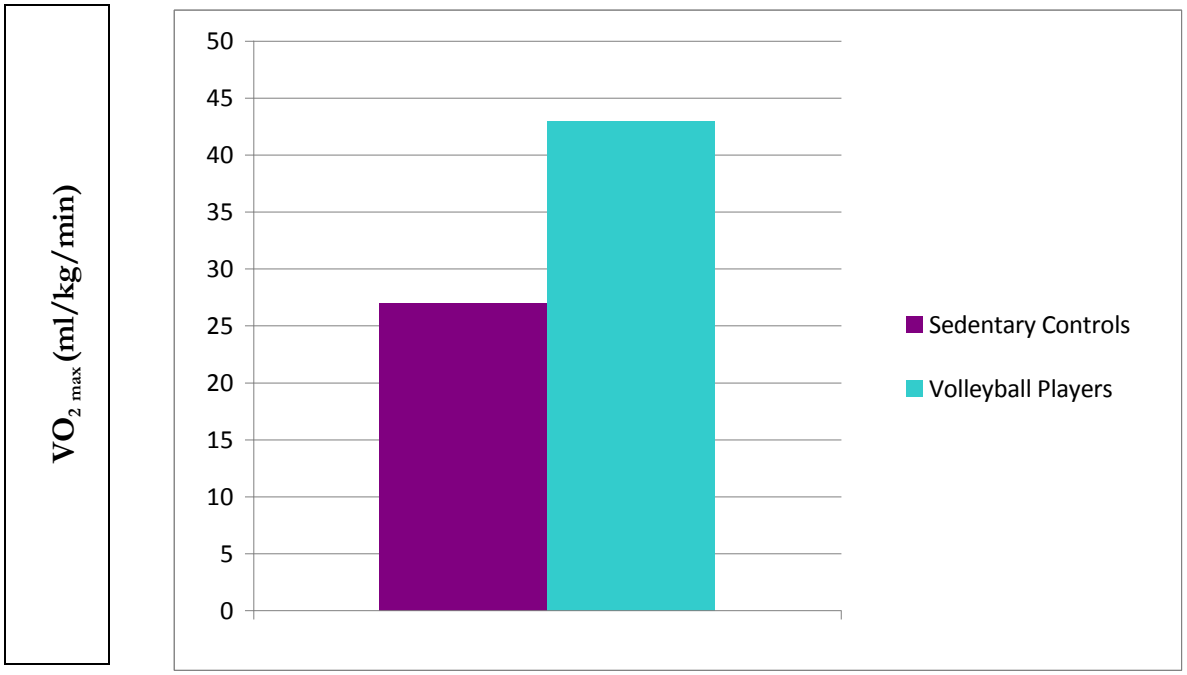
$VO_{2\ max}$ levels of the groups before and after 20 meter shuttle run test were shown in Table 2 and Graphic 1.

Table 2. *Max. Oxygen Uptake ($VO_{2\ max}$ (ml/kg/min))*

$VO_{2\ max}$ (ml/kg/min)	**Sedentary Group n= 10**	**Volleybal Players n= 10**	t	p*
	26.91±3.67	41.78±4.91	13.99	0.000

****significant at p<0.001***

Graphic 1. *Max. Oxygen Uptake ($VO_{2\ max}$ (ml/kg/min))*



After 20 meter shuttle run test, $VO_{2\ max}$ levels of volleyball players were significantly higher than the sedentary controls.

GSH levels of the groups before and after 20 meter shuttle run test were shown in Table 3 and Graphic 2.

n= 10	**Before Test GSH levels(µmol/ml)**	**After Test GSH levels (µmol/ml)**	**t**	**p***
Sedentary Group	17.40±1.25	12.62±1.49	-2.94	0.003
Volleybal Players	17.79±2.12	13.17±1.67	-3.90	0.002

****significant at p<0.05***

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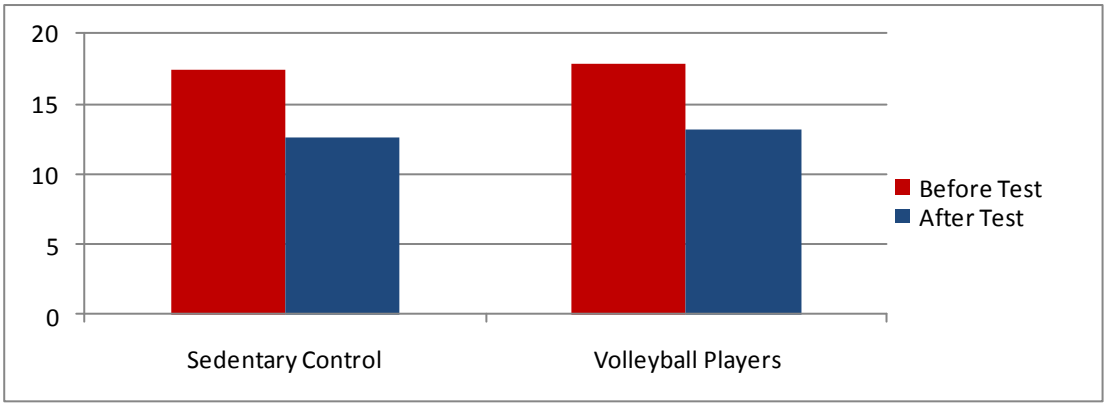
Graphic 2.

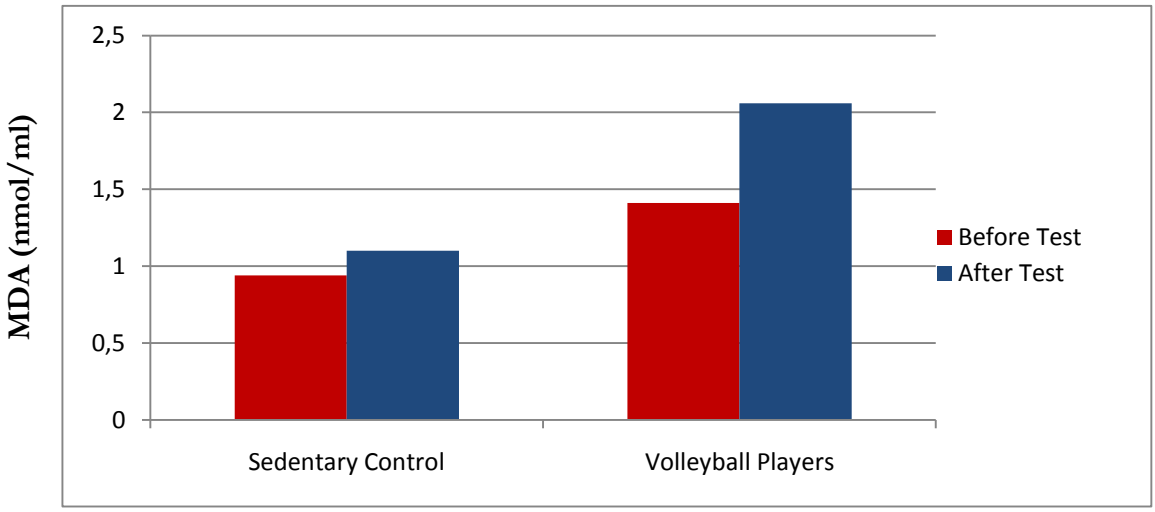


After the test, plasma levels of GSH enzyme levels were found significantly lower than the resting values in both groups ($p<0.05$).

MDA levels of the groups before and after 20 meter shuttle run test were shown in Table 4 and Graphic 3.

n= 10	Before Test MDA levels (nmol/ml)	After Test MDA levels (nmol/ml)	t	p*
Sedentary Group	0.94±0.24	1.10±0.21	-4.59	0.001
Volleyball Players	1.41±0.30	2.06±0.08	-7.47	0.000

significant at p<0.001



After the test, plasma levels of MDA were increased ($p<0.001$) in all subjects.

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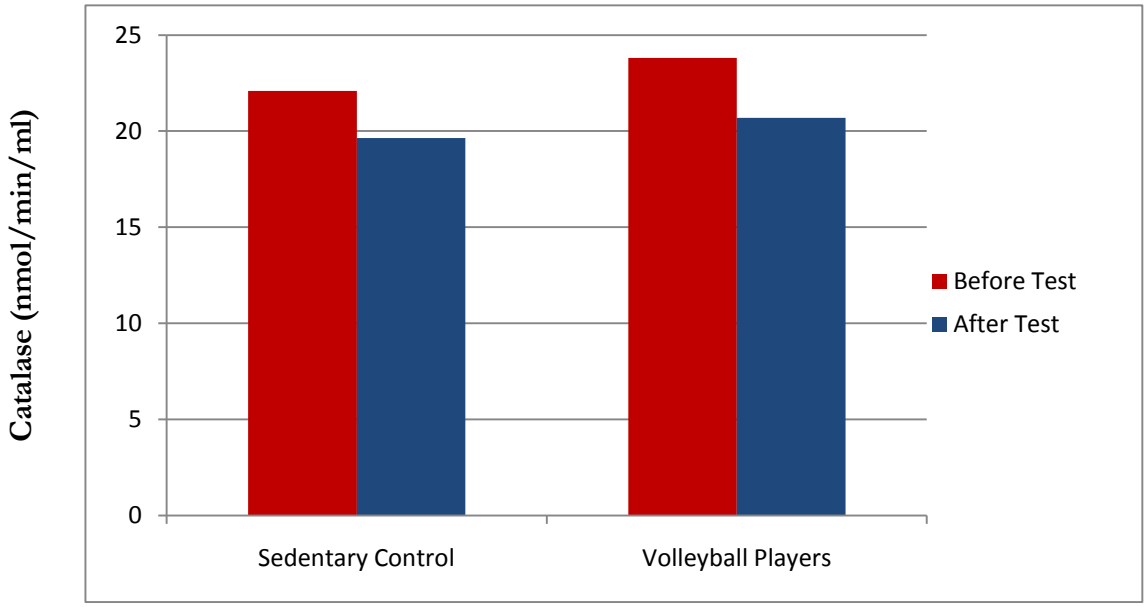
CAT levels of the groups before and after 20 meter shuttle run test were shown in Table 5 and Graphic 4.

n= 10	Before Test Catalase levels (nmol/min/ml)	After Test Catalase levels (nmol/min/ml)	t	p*
Sedentary Group	22.09±2.15	19.64±1.79	5.12	0.001
Volleybal Players	23.80±2.95	20.69±3.42	5.84	0.000

significant at p<0.001



Plasma CAT enzymes levels were found significantly lower than the resting values. Metabolic requirements, increased during exercise, are fulfilled with heart impulse number, pulse volume and blood flow volume. The higher heart pulse volume the higher VO_{2max} is. For the regularly sports performers, they are expected to have higher $VO_{2\ max}$ (Yaprak and Aslan 2008). In our study, $VO_{2\ max}$ values of subjects are compared after exercise test, and $VO_{2\ max}$ values of women volleyball players are found statistically higher than sedentary group.

Free radicals are reacting with macromolecules within the cell, lipids, proteins and DNA, and aiming polyunsaturated fatty acids, frequently found within the structure of the membrane. Systematic oxidation of polyunsaturated fatty acids is called lipid peroxidation.

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Degradation of structural integrity of cell membrane occurs as a result of lipid peroxidation. Especially glucose intake decreases within the cell, and differences are observed on cellular and immunological function responses.

GSH is a tripeptide, which is highly present within various tissues especially in liver, and can be synthesized from glutamate, cysteine and glycine. GSH reacts with free radicals and peroxides, and protect cells against oxidative damages (Öztürk et al. 2001).

GSH has an important role during detoxifications of reactive compounds. GSH is protecting tissues against oxidative damages during exercise. It reduces hydrogen peroxide and organic peroxides. Furthermore GSH reduces tocopherol radicals via reducing semihydroascorbate radical. Thus it forms a protective effect against lipid peroxidation. GSH transmits from plasma to skeletal muscles during exercise (Kurutaş 2001).

In reply to the free radical increase, GSH oxidizes to GSSG. An exhaustive exercise is applied to subjects on treadmill for the purpose of studying the differences at glutation levels after exercise. It is determined that GSSG amount increases within blood after exercise and it returns to its original level within 1 hour. There was no particular change on the blood GSH level (Sastre et al. 1992).

Tessier et al. (1995) determined that blood GSSG increases and GSH/GSSG ratio decreases after maximal aerobic capacity test.

Gohil et al. (1988) notified that blood GSSG increases and GSH levels reduce in connection with elongated submaximal exercise.

Despite these studies, Camus et al. (1994) and Marin et al. (1990) did not determine any changes on blood GSH and GSSG levels after exercise during their studies. This situation shows that term, intense and type of the performed exercise cause various results.

Andersson et al. (2009) determined that plasma GSH/GSSG rate decreases and GSSG rate increases on elite women soccer players as an indicator of oxidative stress at the end of a 90 minute game. This situation is an evidence for increase of oxidative stress production by long term exhausting activity.

Plasma GSH levels, measured on both groups at the beginning of our study, are found logically low at the end of exercise test. ($p < 0.05$)

There is compatibility between findings of performed studies and data of the submitted study.

MDA is one of the most important displays of oxidative stress due to lipid peroxidation. MDA amount increases on various tissues along with the exercise. (Banerjee 2003, Kerksick and Willoughby 2005).

Lekhi et al. (2008) had applied exercise test to 2 groups, namely elite cyclists and sedentary group. They determined that serum MDA and SOD rates of elite cyclists were higher than sedentary group after test. But, serum catalase levels of elite cyclists are found lower than sedentary group at the end of the test. This decrease on catalase activity points the reduce of hydrogen peroxide formation. This situation is an adaptive change for regularly exercising individuals.

Öztaşan et al. (2004) mentioned that acute exhaustive exercise, applied to rats after 8 weeks of endurance workouts, caused increase on erythrocyte MDA levels both for control group and trained group.

Metin et al. (2003), during a study made on soccer players, had determined that plasma MDA levels of soccer players were particularly low compared to control group after Bruce protocol test, where maximal effort is shown.

Bryant et al. (2003), during the study performed on the cyclists, researched the impacts of vitamin C and E intakes on lipid peroxidation connected to exercise. Increase on plasma MDA levels of the subjects, who took only vitamin C, was determined both after resting and exercise. MDA levels of the vitamin E supported group were particularly low compared to placebo users after exercise. At the end of the study, it is determined that vitamin E is more effective against lipid peroxidation compared to vitamin C.

Marini et al. (2007) found that plasma MDA levels of the rats, which are trained for a period of 14 weeks, were higher than control group.

In our study, plasma MDA levels, measured on both groups prior to exercise, logically increased after exercise ($p < 0.001$). Our obtained increased MDA levels are in parallel with the literature.

Düzova et al. (2006) researched the impacts of the intermediate and high level trainings on rats for a period of 13 weeks on treadmill. CAT activities of the rats, which are trained for intermediate term, were found higher than control group. No statistically logical difference is found for the high intensity trained group.

Plasma catalase levels, measured prior to the test as a display of the antioxidant system during our realized study, were found logically higher from the levels, measured after test ($p < 0.001$).

Mignini et al. (2008), at the end of the stress test performed on the athletes, no particular change was determined on plasma MDA concentrations after test but erythrocyte CAT activity was increased with a rate of 33 %. They determined that erythrocyte CAT activity reduced to its original level prior to test after 24 hours.

Cholewa et al. (2008) searched the impacts of Vitamin C intake on blood antioxidant parameters of basketball players after maximal exercise test. No change occurred on the erythrocyte SOD, CAT, GPx and GSH activities of subjects within control group and vitamin support taking group for 21 days after exercise and a low level increase on plasma MDA levels was determined.

Marzatico et al. (1997), during a study on athletes, notified that short distance run did not cause any change on erythrocyte CAT activities but CAT activities were increased on athletes 24 and 48 hours after long distance run.

Rokitzki et al. (1994) mentioned that no change occurred on erythrocyte CAT activities of the athletes, who participated to the marathon, and Aguilo et al. (2000) mentioned that a decrease of 20 % occurred on erythrocyte CAT activities at the end of submaximal exercise, realized on trained cyclists.

Plasma CAT levels, measured prior to the exercise test, had logically decreased after test on both groups ($p < 0.001$). Study of Aguilo et al. shows parallelism with the CAT levels in our study.

In studies, performed on impacts of lipid peroxidation and antioxidant enzymes in connection with exercise, there are conflicting data. In the studies of some researchers, while antioxidant enzyme levels were increasing with the exercise, it does not change on some, and decreases in some other studies. As reasons for getting these kinds of conflicting results, differences on performance of the study on experimental animals or humans, type, intensity and term of the applied exercise, performance format and term of the measurements, and preferred methods.

In our study, when obtained findings are evaluated, they show that 20 meter shuttle - run test stimulates free radical production on both sports women and sedentary group, and also suppresses antioxidant activity.

Conclusions and Recommendations

In our study, as a result;

1) A maximal aerobic test, 20 meter shuttle -run test resulted with increase on free radical production, therefore lipid peroxidation occurred on both sports women and sedentary group.
2) Antioxidant enzyme activities are found particularly low on both groups compared to resting levels. Probably this event could be due to two factors.

a) Free radicals cause lipid peroxidation, and change membrane structure and viscosity, and thus cause change on antioxidant status.
b) Although formation free radicals increase in connection with the oxidative stress, induced with high intensity exercise, antioxidant capacity can be insufficient for responding to this increase.

Therefore, due to increase of oxidative stress and accordingly occurred free radical damages by acute exhaustive exercises, regular exercises without high intensity and are recommended in connection with physical health.

References

- Aguiló A, Tauler P, Gimeno I, Fuentespina E, Pons A. (2000). Changes in erythrocyte antioxidant enzymes during prolonged submaximal exercise. Biofactors.11(1-2):27-30.
- Alessio HM, Hagerman AE, Fulkerson BK, Ambrose J, Rice RE, Wiley RL. (2000). Generation of reactive oxygen species after exhaustive aerobic and isometric exercise. Med Sci Sports Exerc. Sep;32(9):1576-81.
- Andersson H, Karlsen A, Blomhoff R, Raastad T, Kadi F. (2009). Plasma antioxidant responses and oxidative stress following a soccer game in elite female players. Scand J Med Sci Sports. 2009. 35-42.
- Banerjee AK, Mandal A, Chanda D, Chakraborti S. (2003). Oxidant, antioxidant and physical exercise. Mol Cell Biochem. Nov;253(1-2):307-12.

- Brites FD, Evelson PA, Christiansen MG, Nicol MF , Basilico MJ, Wikinski RW, Llesuy SF. (1999). Soccer players under regular training show oxidative stress but an improve plasma antioxidant status. Clinical Science 96:381–385.
- Bryant RJ, Ryder J, Martino P, Kim J, Craig BW. (2003). Effects of vitamin E and C supplementation either alone or in combination on exercise-induced lipid peroxidation in trained cyclists. J Strength Cond Res. Nov;17(4):792-800.
- Camus G, Felekidis A, Pincemail J, Deby-Dupont G, Deby C, Juchmes-Ferir A, Lejeune R, Lamy M. (1994). Blood levels of reduced/oxidized glutathione and plasma concentration of ascorbic acid during eccentric and concentric exercises of similar energy cost. Arch Int Physiol Biochim Biophys. Jan-Feb;102(1):67-70.
- Cholewa J, Poprzęcki S, Zajac A, Waskiewicz Z. (2008). The influence of vitamin C on blood oxidative stress parameters in basketball players in response to maximal exercise.Science&Sports.23;(3-4):176-82.
- Clarkson PM, Thompson HS. (2000). Antioxidants: what role do they play in physical activity and health?. Am J Clin Nutr. Aug;72(2 Suppl):637S-46S.
- Deaton CM, Marlin DJ. (2003). Exercise-associated oxidative stress. Clin Tech Equine Prac. 2(3):278-291.
- Demirbağ R, Yilmaz R, Güzel S, Celik H, Koçyigit A, Ozcan E. (2006). Effects of treadmill exercise test on oxidative/antioxidative parameters and DNA damage. Anadolu Kardiyol Derg. Jun;6(2):135-40.
- Dinçer C. (2001). Egzersizle oluşan kas hasarında serbest radikal mekanizmalar. Spor ve Tıp. 9(7-8):11-14.
- Dündar Y, Aslan R. (1999). Hücre moleküler statüsünün anlaşılması ve fizyolojik önem açısından radikaller-antioksidanlar. Cerrahi Tıp Bilim. Derg. 2(2):134-142.
- Düzova, H, Emre MH, Karakoç Y, Karabulut AB, Yılmaz, Z, Gürsul C, Yoloğlu S. (2006). Orta ve yüksek düzeyde treadmill egzersizinin sıçanların kas ve eritrosit oksidan ve antioksidan sistemine etkisi. İnönü Üniversitesi Tıp Fakültesi Dergisi. 13(1): 1-5.

- Fıçıcılar H, Zergeroglu AM, Tekin D, Ersoz G. (2003). The effects of acute exercise on plasma antioxidant status and platelet response. Thromb Res. 111(4-5):267-71.
- Gohil K, Viguie C, Stanley WC, Brooks GA, Packer L. (1988). Blood glutathione oxidation during human exercise. J Appl Physiol. Jan; 64(1):115-9.
- Kara M, Gökbel H. (1997). Maksimal aerobik gücü etkileyen faktörler. Genel Tıp Dergisi. 7(1):39-42.
- Kerksick C, Willoughby D. (2005). The antioxidant role of glutathione and N-acetyl-cysteine supplements and exercise-induced oxidative stress. J Int Soc Sports Nutr. Dec 9;2:38-44.
- Kılıç M, Baltacı AK, Gunay M, Gökbel H, Okudan N, Cicioglu I. (2006). The effect of exhaustion exercise on thyroid hormones and testosterone levels of elite athletes receiving oral zinc. Neuro Endocrinol Lett. Feb-Apr;27(1-2):247-52.
- Kurutaş E. (2001). Endosülfanın fare eritrosit antioksidan sistemleri ve malondialdehit düzeyleri üzerine etkisi. Çukurova Üniversitesi Tıp Fakültesi Dergisi, 26(1):14-19.
- Lambertucci RH, Levada-Pires AC, Rossoni LV, Curi R, Pithon-Curi TC. (2007). Effects of aerobic exercise training on antioxidant enzyme activities and mRNA levels in soleus muscle from young and aged rats. Mech Ageing Dev. Mar; 128(3):267-75
- Lamprecht M, Greilberger J, Oettl K. (2004). Analytical aspects of oxidatively modified substances in sports and exercises. Nutrition. Jul-Aug;20(7-8):728-30.
- Lekhi C, Gupta PH, Singh B. (2008). Influence of exercise on oxidant stress products in elite Indian cyclists. Br J Sports Med. Oct;41(10):691-3.
- Limón-Pacheco J, Gonsebatt ME. (2009). The role of antioxidants and antioxidant-related enzymes in protective responses to environmentally induced oxidative stress. Mutat Res. Mar 31;674(1-2):137-47.
- Marin E, Hänninen O, Müller D, Klinger W. (1990). Influence of acute physical exercise on glutathione and lipid peroxides in blood of rat and man. Acta Physiol Hung. 76(1):71-6.

- Marini M, Lapalombella R, Margonato V, Ronchi R, Samaja M, Scapin C, Gorza L, Maraldi T, Carinci P, Ventura C, Veicsteinas A. (2007). Mild exercise training, cardioprotection and stress genes profile. Eur J Appl Physiol. Mar;99(5):503-10.
- Marzatico F, Pansarasa O, Bertorelli L, Somenzini L, Della Valle G. (1997). Blood free radical antioxidant enzymes and lipid peroxides following long-distance and lactacidemic performances in highly trained aerobic and sprint athletes. J Sports Med Phys Fitness. Dec;37(4):235-9.
- Metin G, Gümüştaş MK, Uslu E, Belce A, Kayserilioglu A. (2003). Effect of regular training on plasma thiols, malondialdehyde and carnitine concentrations in young soccer players. Chin J Physiol. Mar 31;46(1):35-9.
- Mignini F, Tomassoni D, Traini E, Streccioni V. (2008). Antioxidant endogenous defense in a human model of physical stress. Clin Exp Hypertens. Nov;30(8):776-84.
- Özçelik O, Karataş F. (2008). Şiddeti düzenli olarak artan işe karşı yapılan egzersizin obezlerde serum malondialdehid ve vitamin A, E, C düzeyleri üzerine olan etkisi. F.Ü. Sağ. Bil. Derg. 22 (6): 337–341.
- Özdemir O. (2005). Medikal İstatistik. 1.Baskı. İstanbul, istanbul Medikal Yayıncılık, 130-138.
- Öztaşan N, Taysi S, Gumustekin K, Altinkaynak K, Aktas O, Timur H, Siktar E, Keles S, Akar S, Akcay F, Dane S, Gul M. (2004). Endurance training attenuates exercise-induced oxidative stress in erythrocytes in rat. Eur J Appl Physiol. May;91(5-6):622-7.
- Öztürk M, Güzelhan Y, Sayar K, Tüzün Ü. (2001). Yaygın Gelişimsel Bozukluğu olan Çocuklarda Plazma Malondialdehit ve Glutatyon Düzeylerinin Araştırılması. Bull Clin Psychopharmacol, 11:155-159.
- Ramsbottom R, Brewer J, Williams C. (1988). A progressive shuttle run test to estimate maximal oxygen uptake. Br J Sports Med. Dec;22(4):141-4.
- Rokitzki L, Logemann E, Sagredos AN, Murphy M, Wetzel-Roth W, Keul J. (1994). Lipid peroxidation and antioxidative vitamins under extreme endurance stress. Acta Physiol Scand. Jun;151(2):149-58.

- Sacheck JM, Blumberg JB. (2001). Role of vitamin E and oxidative stress in exercise. Nutrition, Oct;17(10):809-14.
- Sastre J, Asensi M, Gascó E, Pallardó FV, Ferrero JA, Furukawa T, Viña J. (1992). Exhaustive physical exercise causes oxidation of glutathione status in blood: prevention by antioxidant administration. Am J Physiol. Nov;263(5 Pt 2):R992-5.
- Sınırkavak G, Dal U, Çetinkaya Ö. (2004). Elit sporcularda vücut kompozisyonu ile maksimal oksijen kapasitesi arasındaki ilişki. C.Ü. Tıp Fakültesi Dergisi. 26 (4): 171-176.
- Tessier F, Margaritis I, Richard MJ, Moynot C, Marconnet P. (1995). Selenium and training effects on the glutathione system and aerobic performance. Med Sci Sports Exerc. Mar;27(3):390-6.
- Valko M, Leibfritz D, Moncol J, Cronin MT, Mazur M, Telser J. (2007). Free radicals and antioxidants in normal physiological functions and human disease. Int J Biochem Cell Biol. 39(1):44-84.
- Viña J, Gomez-Cabrera MC, Lloret A, Marquez R, Miñana JB, Pallardó FV, Sastre J. (2000). Free radicals in exhaustive physical exercise: mechanism of production, and protection by antioxidants. IUBMB Life. Oct-Nov;50 (4-5):271-7.
- Yaprak Y, Aslan A. (2008). Üniversite badminton takımı oyuncularının kalp debisi, VO_2max ve solunum fonksiyon testlerinin karşılaştırılması. Spormetre Beden Eğitimi ve Spor Bilimleri Dergisi. VI (2): 69-74.