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The Effects of Vitamin E Application on Some Free Radicals and Lactate Levels in Acute Exercise

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ABSTRACT

The aim of this study is to investigate the effect of vitamin E supplement on elite taekwondo performers who were made to do acute taekwondo exercise on some free radicals. Twenty-four healthy male elite taekwondo performers whose mean age is 21.66 ± 1.20 and mean body weight is 72.50 ±8.17 kg participated the study. The study protocol was also endorsed by the ethical board of the same faculty. In the study, the subjects were divided into 2 equal groups. The vitamin E group (Group 1) will be given vitamin E for 4 weeks (300 mg D-alpha- tocopheryl acetate) and will be made to do acute taekwondo exercise once a week till they get tired. The control group (Group 2) is the group who were only made to do acute taekwondo exercise till they got tired once a week for 4 weeks. Taekwondo exercises were carried out for four weeks and once a week as acute exercises till they got tired. During four weeks of application procedure, malondialdehyde (MDA) and Nitrococsite (NO) levels and plasma lactate (LAC) levels were determined from the blood samples taken from the subjects before the beginning of and at the end of the application and before and after the training. While the highest MDA level was obtained from the 2nd group in the 4th measurement ($P<0.05$), the lowest MDA level was obtained from 3rd measurement in the 1st group ($P<0.05$). While the highest NO level was obtained in the 2nd measurement in the 2nd group ($P<0.05$), the lowest NO level was obtained on the 3rd measurement in the 1st group ($P<0.05$). While the highest lactate level was obtained in the 2nd measurement in group 1 ($P<0.05$), the lowest lactate level was in the 3rd measurement in group 1 ($P<0.05$). As a result, it was determined that vitamin E application prevented free radical formation by increasing antioxidant activity during acute taekwondo exercise and substantially pressurized lactate levels.

Keywords: Acute exercise, vitamin E, free radicals, lactate.

INTRODUCTION

Human body has a certain capacity to adapt to various inner and outer sources of stress (19). Physical exercise can be regarded a source of such stress. During exercises, oxidative stress which develops in parallel to increasing oxygen consumption fastens free radical production. Acute exercise leads to oxidative stress, damage to muscle structure, lipid peroxidation in the membranes and free radical spectrum. In damaged tissue, phospholipids lead to proteinase enzyme activation and cell membrane and arachidonic acid release in cell membrane, which in turn lead to increase in oxidation and free radical production (3). Free radical generation resulting from exercise and lipid peroxidation caused by this is one of the damaging mechanisms for organism (1,8,19).

The amount of oxygen consumed during exercise varies according to the intensity and the type of the exercise; it can generally increase up to 10-15 times compared to rest (1,19). Considering that there are thousands of molecules of free radical production at rest, it is natural to expect that free radical production will rise significantly during exercise when metabolism is highly fast.

In most of the studies, it is known that acute sub-maximal exercise lead to lipid peroxidation and that regular exercise lead to positive change in antioxidant status. In trained people, the existence of stable oxidative stress both in muscle cells and in other cells lead to the formation of a strong antioxidant system to protect itself from risk which can result from exercise. However, protection from excessive oxidative stress which can result from unprepared exercise is difficult. Studies show that programmed physical activity develop the antioxidant status of blood dependent on chronic oxidative stress, thus it plays a role in the formation of a strong antioxidant mechanisms (3).

Vitamin E is an important membrane stabilizer settled on the cell membrane which plays a role in the antioxidant defense with its antioxidant property. It is taken by sportsmen very commonly, especially in the last 40 years, to prevent damage to muscle resulting from exercise and to fasten pulling together after exercise (32).

The results of animal and human studies so far indicate that vitamin E plays a great role in doing away with free radicals, which result from exercise, and their hazardous effects (6,17). When the effect of vitamin E on performance is examined under the light of existent sources, it is seen that the study of Simon-Schnass and Past (31) on climbers reports obvious positive and the study by Novelli et al. (25) on mice in which they examined swimming endurance performance. However, when some complied literature is examined, it is seen that vitamin E does not have a positive effect on exercise (19,32). Following from these, this study aims to study the effect of vitamin E supplement on elite taekwondo performers who do acute taekwondo exercise.

The MDA and NO levels and plasma lactate levels were determined in the blood samples taken from the subjects and before and after the training before and after the application period which lasted four weeks.

MATERIAL & METHOD

Method

This study was carried out on 24 healthy male students at School of Physical Education and Sports at Selcuk University. Their mean age was 21.66 ± 1.20 years and mean body weights was 72.50 ±8.17 kg. The subjects were chosen among elite sportsmen who are students attending School of Physical Education and Sports at S.U. and actively performing taekwondo. The study protocol was endorsed by the ethical board of School of Physical Education and Sports at Selçuk University on 11.02.2008 with issue number 2008/001.

The subjects were divided into 2 groups each having the same number of members.

Vitamin E (Group 1) group: This is the group which was given vitamin E (300 mg alfa-tocopherol acetate) for 4 weeks and who were made to do taekwondo exercise till they got tired.

Control (Group 2) group: The control group which did acute taekwondo exercise till they got tired once a week for 4 weeks.

Taekwondo exercises were done for 4 weeks as acute exercises till they got tired and they exercised in pairs.

Experimental Applications

Vitamin E Application

Vitamin E was given orally as tablet (300 mg alpha-tocopherol acetate) at 9 o'clock during four weeks period.

Acute Taekwondo Exercise

The groups were made to do acute Taekwondo exercise once in a week during four weeks. The exercises began with general heating for 20 minutes. After heating, the players were taken into gloves study one by one. The players applied all techniques on gloves until exhausted by maximal overloading through using all taekwondo techniques. This study was repeated in 3 sets.

Biochemical Analyses

The blood samples that were received from elbow vein in due form were poured into tubes including Ethylenediaminetetraacetic acid (EDTA) and then immediately applied centrifuge at +4°C degree 3500 rpm for 15 minutes and then plasma and serum were availed.; Melandialdehid (MDA) Assay kit, Cayman mark (NO) Assay kit from the plasma and serum samples were used and MDA and NO levels were measured by radio immunoassay and Elisa methods by using commercial kits.

Plasma Lactate Assignments

Sufficient blood that was received from ear lobe was assigned by lactate analyzer (VARIO Photometer, Germany). Plasma lactate levels were assigned as (through reading 550 nm wave lengths) mg/ dl. The results were determined as U/ml.

Statistical Evaluations

The statistical analysis was performed with use of the SPSS 10.3 software. The data were presented as mean and standard deviation (Mean ± SD). Mann-Whitney U test was used for differences between parameters in the statistical analyses. Level of significance was set at $p<0.05$.

RESULTS

In this study, the groups are as follows;

Vitamin E (Group 1) Group: This group which was applied acute Taekwondo exercise until fatigue feeling once in a week period was given orally Vitamin E (300 mg alpha-tocopherol acetate) during four weeks period..

Control (Group 2) Group: The control group which was applied acute taekwondo exercise until they felt fatigue once in a week during 4 weeks period.

When intergroup MDA level in the study was examined (table 1; figure 1), the highest and the most important MDA level was availed from timing of Control (Group 2) Group's fatigue timing after sustenance ($p<0.05$). No any important difference at intergroup MDA level was found from other timings of both groups ($p>0.05$). When MDA levels of Vitamin E (Group 1) group was compared at the study, the highest MDA value was determined at the 2nd (fatigue before sustenance) measuring ($p<0.05$). While the fatigue after sustenance was found higher in important degree than (4th measuring) MDA value, 1st (resting before sustenance) and the 3rd (fatigue after sustenance) measuring value ($p<0.05$); the 1st (resting before sustenance) and 3rd (resting after sustenance) MDA measuring values were found indifferent from each others ($p>0.05$). When the MDA level of Control (Group 2) group was evaluated; the highest MDA level was availed at the 4th (fatigue after sustenance) measuring. While the 4th (fatigue after sustenance) measuring value was similar with the 2nd (fatigue before sustenance) measuring value ($p>0.05$), 2nd (fatigue before sustenance) measuring value was found higher than other measuring groups in important level ($p<0.05$). The 1st and 3rd measuring levels were indifferent from each others ($p>0.05$).

When intergroup NO level in the study was examined (table 2; figure 2), NO level in the 4th (fatigue after sustenance) measuring value in the Control (Group 2) was found higher and more important level than Vitamin E (group 1) Group ($p<0.05$). No any difference was determined among other measuring timings ($p>0.05$). When Vitamin E (Group 1) and Control Group (group 2) NO levels were compared at the study, the highest NO value was determined at the 2nd (fatigue before sustenance) ($p<0.05$). The 4th (fatigue after sustenance) measuring NO value was found higher than 1st (Resting before sustenance) and 3rd (Resting after sustenance) measuring values in important level ($p<0.05$); but the 1st Resting before sustenance) and the 3rd (Resting after sustenance) measuring NO values were similar to each others ($p>0.05$).

Table 1. Plasma MDA Levels (nmol/ml) of Working groups

Group (n=12)	MDA Before Sustenance		MDA After Sustenance	
	Resting (1st meas.)	Fatigue (2nd meas.)	Rest (3rd meas.)	Fatigue (4th meas.)
Vitamin E(Group1)	4.84 ± 0.35^{c}	9.08 ± 0.52^{a}	4.74 ± 0.14^{c}	7.18 ± 0.50^{Bb}
Control (Group 2)	5.80 ± 0.36^{b}	10.60 ± 0.51^{a}	5.94 ± 0.38^{b}	11.00 ± 1.04^{Aa}

a, b, c: The difference between averages including different letters at the same line is statistically important (P< 0,05).
A,B: The difference between averages including different letters at the same column is statistically important (P< 0,05).

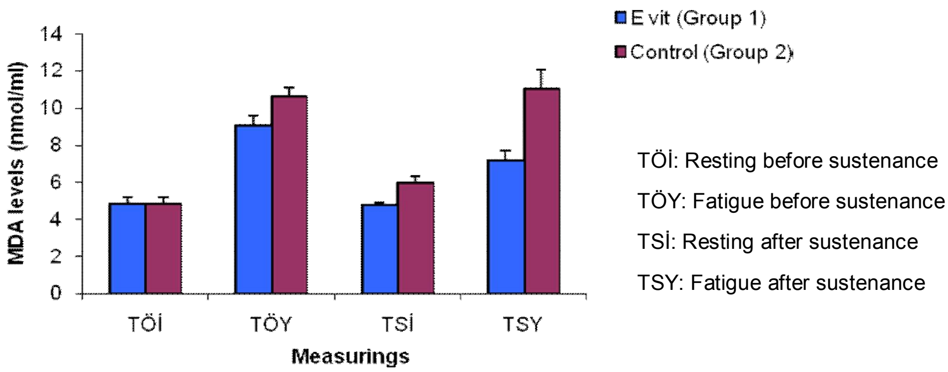

Figure 1. MDA Levels of the Groups (nmol/ml).

Table 2. Serum NO Levels (μM) of Working Groups

Group (n=12)	NO Before Sustenance		NO After Sustenance	
	Resting (1st meas.)	Fatigue (2nd meas.)	Rest (3rd meas.)	Fatigue (4th meas.)
Vitamin E (Group 1)	6.32 ± 0.34^{c}	11.94 ± 0.98^{a}	5.38 ± 0.30^{c}	9.14 ± 0.28^{Bb}
Control (Group 2)	5.50 ± 0.34^{c}	12.48 ± 0.60^{a}	5.44 ± 0.65^{c}	10.84 ± 0.56^{Ab}

a, b, c: The difference between averages including different letters at the same line is statistically important (P< 0,05).
A,B: The difference between averages including different letters at the same column is statistically important (P< 0,05).

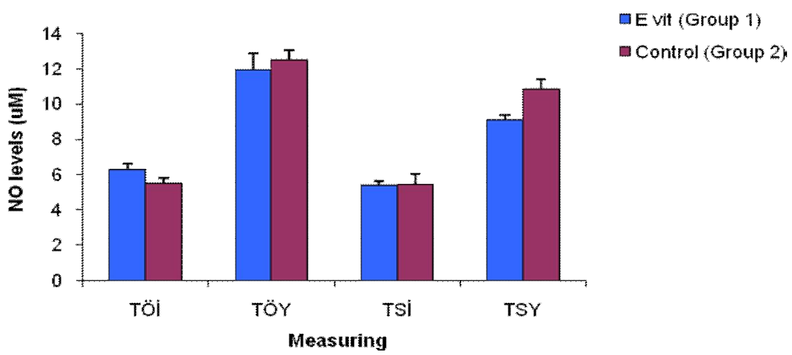

Figure 2. NO values of the Groups (μM).

When intergroup Lactate levels in the study was examined (table 3; figure 3), Lactate level in the 2nd (fatigue before sustenance) measuring value in the Vitamin E (group 1) was found higher and more important level than Control Group ($p<0.05$); the 3rd (Resting after sustenance) measuring value was found lower in important degree than control (Group 2) group ($p<0.05$). Other measuring timing is similar to each others ($p>0.05$). When Lactate level of Vitamin E (Group 1) group was examined at the study, the highest Lactate level was evaluated from the 2nd (fatigue before sustenance) measuring ($p<0.05$). While the 4th (Fatigue after sustenance) measuring Lactate value was found higher than the 1st (Resting before sustenance) and the 3rd (Resting after sustenance) measuring values in important level ($p<0.05$); the 1st (Resting before sustenance) and the 3rd (Resting after sustenance) measuring Lactate values are similar to each others ($p>0.05$). When the Lactate levels of Control group (Group2) were compared; the highest Lactate value was evaluated from the 2nd measuring ($p<0.05$). While the 2nd (Fatigue before sustenance) measuring value was similar to the 4th (Fatigue after sustenance) measuring value ($p>0.05$), other measuring timing was found high in important level ($p<0.05$). The LAC measuring level of the 1st (Resting before sustenance) and the 3rd (Resting after sustenance) measuring levels were similar to each others.

DISCUSSION

Discussion of MDA Findings

When the MDA Levels before and after subsistence of both groups were examined at the study, it has been seen that, fatigue MDA levels increased in important degree than resting MDA levels. Although it is known that regular physical activity is important for health, it is remarked that the exercises that are performed temporarily (acutely) or in high fatigue may increase oxidative damage may increase due to ROT (Reactive Oxygen Types) (36). Davison et al. (7) showed that strength trainings increased DNA damage although applying antioxidant materials. Although there is conflict information, consequently it is agreed that physical exercise increases free radical composing (20). Beside heavy exercise can increase all body oxygen intake 20 times more, can increase oxygen consumption in the active muscle fibers 200 times more than resting level (5). At the end of those developing events, it is stated that existing mitochondrial metabolic leaks causes increasing at free radical molecule producing during exercises (15). It is propounded that the intense physical activities at not only human but also animal causes oxidative damage at blood and various tissue (11,26). No any important difference was found at the resting values before and after subsistence of both groups at the study. This can be explained as no any composing free radical occurred at the metabolism during resting; so no any important change occurred in MDA levels.

Table 3. Lactate (LAC) Levels (mmol/l) of Working Groups

Group (n=12)	LAC Before Sustenance		LAC After Sustenance	
	Resting (1st meas.)	Fatigue (2nd meas.)	Rest (3rd meas.)	Fatigue (4th meas.)
Vitamin E (Group 1)	1.12 ± 0.55^{c}	11.58 ± 1.08^{Aa}	1.10 ± 0.30^{Bc}	8.69 ± 1.24^{b}
Control (Group 2)	1.16 ± 0.33^{b}	9.14 ± 1.71^{Ba}	1.57 ± 0.54^{Ab}	8.49 ± 1.00^{a}

a, b, c: The difference between averages including different letters at the same line is statistically important (P< 0,05).
A,B: The difference between averages including different letters at the same column is statistically important (P< 0,05).

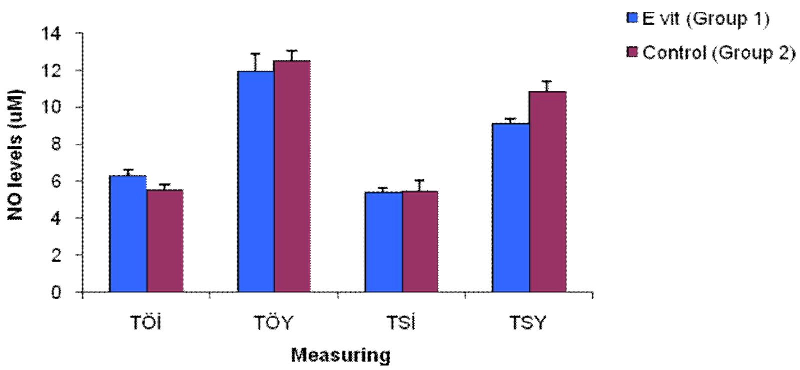

Figure 3. LAC Levels of the Groups (mmol/l).

When MDA levels of Vitamin E Group were compared in the study, it has been stated that the highest MDA value was determined at the 2nd (Fatigue before sustenance) measuring. MDA level of fatigue after sustenance (4th measurement) decreased significantly compared to MDA level of fatigue before sustenance (2nd measurement). Besides, fatigue after sustenance value (4th measurement) of Vitamin E group (Group 1) decreased significantly compared to the control group (group 2). It was determined by Kaczmarski et al. (18) that MDA production during physical exercise is pressed through combine application of vitamin E, selenium, vitamin C. Similarly, it has been set forth that selenium and vitamin E application for 6 weeks at the diabetic rats which were also applied exercises decreased MDA levels (21). As it is mentioned in another literature, at the end of MDA analyzes after maximum exercise application of people who were applied 300 mg/day Vitamin E for 4weeks, it has been stated that lipid oxidation based to exercise has been decreased (10). The reports of mentioned researchers are important as they show that Vitamin E application decreased increasing MDA producing and it shows similarity with our findings.

Not determining an important difference between MDA values of both groups at the resting before sustenance (1st measuring) and fatigue (2nd measuring) and Resting after sustenance (3rd measuring) make us thinking that oxidative action does not actuate during resting so it doesn't affect MDA levels and both groups experimental subjects have similar physical performance to each other in the study.

Discussion of NO Levels

When NO level after and before sustenance of both groups is compared in the study, it has been seen that fatigue NO level increased in more importantly degree than resting levels. Blood stream at the exercise cannot meet increasing oxygen consuming at the tissues (9). Insufficient or decreasing blood stream in the organism increases NO producing. On the other hand, xanthin that is resource of superoxide anion resource turns into dehydrogenize during ischemia (16,29). So, SOD activity and nitric oxide level may show parallelism with medium and high level exercise. On the other hand, existing negative correlation between CAT activity and NO in medium level exercise can be a result of increasing antioxidant capacity and/or a result of increasing muscle tissue superoxide dismutase enzyme activity (8). Güllü (14) determined that there was important increasing at NO levels between before exercise and after exercise at his implemented study over sportsmen and sedentary individuals. Also, Goto et al. (12) states that regular chronic exercise increases NO level and causes vasodilatation. The knowledge that availed at the literature shows similarity with our study. When intergroup fatigue after sustenance NO values were compared in our study, fatigue value of Vitamin E (group 1) group (4th measuring) decreased in important level more than Control group (group 2). In accordance with statement by Minamiyama et al. (24), Vitamin E additive caused oxidative stress at rats, rapidly tolerated NO increasing. Also Ushiyama et al. (35) stated that applying vitamin E sustenance to hypertension patients for 8 weeks and caused high blood pressure but it decreased increasing NO level to important level and this shows similarity with findings that we availed in our study.

No any important difference was found between intergroup resting before sustenance and fatigue (1st and 2nd measuring) NO values and Resting after sustenance (3rd measuring). This show that not increasing oxidative stress during resting so occurring no any important change at NO level and experimental subjects of both groups have similar physical performance to each others. Traverse et al. (2000)'s stating that not changing NO level even at the low degree exercise supports our findings.

Discussion of Lactate Levels

When lactate level after and before sustenance of both groups is compared in the study, it has been seen that fatigue lactate level increased in more importantly degree than resting levels. Also it is stated that lactate concentration was increased in plasma against heavy exercise (13). In accordance with statement by Grant et al. (13), Rodas et al. (2000), lactate concentration was higher during long term and heavy exercise and this shows similarity with findings that we availed high lactate levels during acute taekwondo exercise in our study. Fatigue after sustenance value of Vitamin E group decreased in important degree pursuant to level of fatigue before sustenance. Beside this, fatigue after sustenance value of Vitamin E group is similar with fatigue before/after sustenance value of control group and this result can be explained by the idea that Vitamin E suppresses lactate level significantly. In accordance with statement by Tsakiris et al. (34), 200 mg. Vitamin E supplementation decreased lactate level. In a similar way, in accordance with statement by Chae et al. (4) and Aquilo et al. (2), Vitamin E supplementation decreased lactate level and it shows similarity with our findings.

Resting after sustenance value of Vitamin E group decreased in important degree pursuant to resting value of the other group and this can be explained by Vitamin E effect as reported by Machefer et al. (23) and Schulpis et al. (30). Beside this, in accordance with statement by Koca et al. (22), Fatigue before sustenance value of control group decreased in important degree pursuant to Vitamin E group and it shows that Exercise Group physical training is better than Control Group. No any important difference was found at the resting values before subsistence of both groups. This can be explained as lactate levels were not agitated during resting. Also fatigue after sustenance value is similar for the both groups. This can be explained by the effect of Vitamin E supplementation for 4 weeks (4,2).

As a result, it was determined that vitamin E application prevented free radical formation by increasing antioxidant activity during acute taekwondo exercise and substantially pressurized lactate levels.

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