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Effect of 2 Weeks Vitamin E Supplementation to the Point of Anaerobic Threshold

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ABSTRACT The purpose of this study was to examine the effects of 400 mg vitamin E supplementation each day for two weeks on anaerobic threshold level. The study was done on volunteered male subjects (n=30) who were between the ages of 19-24 and they were students at the department of physical education and sports. In this study, Conconi test was used to determine the Anaerobic Threshold (AT) of the subjects and the True 750 S.O.F.T System treadmill was used to determine the AT and training intensity. Polar Heart Rate monitor was used to determine the heart rates of the subjects. Also, Roche Accutrend Lactate Analyser was used to determine the Blood Lactic Acid (BL) levels of the subjects. The subjects were divided randomly into three groups (vitamin E, placebo, and control). The subjects who were in the vitamin E and placebo groups took either vitamin E or placebo for two weeks. All measurements were taken before and after the supplementation period. Wilcoxon and Kruskal-Wallis methods were used to evaluate the effects of supplementation ($p<0.05$). The results of the study showed that there was no significant difference among the groups in all the parameters [Heart Rate at Anaerobic Threshold (ATHR), Running Speed at Anaerobic Threshold (ATRS), Blood Lactate at Anaerobic Threshold (ATBL), Maximal Running Distance (RDM), and Maximal Heart Rate (HRM)]. From these results, it may be concluded that the daily intake of 400 mg vitamin E for two weeks had no effect on the physical performance of this group.

INTRODUCTION

Severe acute physical exercise causes a lot of oxygen consumption and thus leads to free radical formation (Wotton 1988). Even with low intensity exercise for strength training, there is lipid peroxidation (Cakir-Atabek et al. 2015). Increased physical activity and muscle contractions during exercise, causes energy consumption and metabolic activities to accelerate significantly. During exercise, oxygen consumption increases by 10-40 times and this increased oxygen consumption causes free radical production (Silbernagl and Despopoulos 1989; Sen 2001). Free oxygen radicals are extremely chemically active substances. Numerous studies show that free oxygen radicals cause deformation in various tissues such as the cell membrane and nucleic acids (Zergeroglu et al. 1997). Antioxidants prevent oxidation by reacting with free radicals in body tissues. There are some vitamins, minerals and enzymes that have antioxidant effects in the human body; Vitamin E, vitamin C and vitamin A, which are antioxidant vitamins (Ersoy 2000). Vitamin E is likely the most important antioxidant in human nutrition (Wallert et al. 2014). Some researches show that antioxidants, especially vitamin E, protect the body from free radical damage caused by heavy exercise and improve exercise capacity (Ersoy and Hasbay 1998; Vina et al. 2000; Leeuwenburgh and Heinecke 2001; Mondal and Bandopadhyay 2016). In healthy individuals, increasing vitamin E intake up to 2-10 times (20-100 mg / day) was found to improve immune functions and vitamin E supplementation, an important factor in the defense against oxidative stress and muscle damage (Silva et al. 2010; Santos et al. 2016). But some other studies indicate that antioxidant vitamins have had no effect on oxidative stress and exercise performance (Mohammed et al. 2015; Morrison et al. 2015). However, it is stated that suggesting the use of antioxidants precisely for individuals who exercise, need to have more study (Peternelj and Coombes 2011).

There are no generally accepted dietary recommendations about vitamin E. However, up to 300 mg daily use of vitamin E does not generate any physiological problem (Schmölz et al. 2016). Is has also been reported that the use of more than 1 gram of vitamin E per day, causes some clinical problems such as hypertension, fatigue, breast tumors, muscle weakness, vision problems, reproductive disorders and delayed wound healing (Dunne 1990; Ersoy and Hasbay 1998).

Anaerobic threshold (AT) is considered as one of the most important indicators of endurance capacity. Many research studies have indicated that endurance trainings have limited contribution to the development of maximal oxygen uptake capacity (VO_2 max%). However, they noted that anaerobic threshold trainings have more important contributions to the development of endurance capacity (Atalay et al. 1997). The anaerobic threshold is defined as the level of exercise intensity at which lactic acid builds up in the body faster than it can be cleared away (Sonmez 2002). Anaerobic threshold is found in about eighty percent of maximal heart rate values, that is 150-170 beats/minutes. This value may decline to 130 beats/min for untrained individuals, and rise to 180 beats/minutes for trained individuals (Gur 1992). Although the maximal oxygen uptake capacity, which is an important criterion in terms of aerobic endurance is due to genetic factors (between 80-90%), anaerobic threshold point depends entirely on trainings. Sedentary individuals' anaerobic threshold point occurred at 50-60 percent of VO_2 max, whereas, in trained individuals, the anaerobic threshold point can reach 80-90 percent of VO_2 max (Davis 1985).

An improvement in the anaerobic threshold; helps to increase the capacity for work, reduces lactic acid build up in working muscles and help faster removal of lactic acid from the muscles. However, the effects of antioxidant vitamin supplementation on exercise performance and oxidative stress in skeletal muscle is still uncertain (Conti et al. 2016; Merry and Ristow 2016). Therefore, the aim of this study was to investigate the effect of vitamin E supplementation to the point of anaerobic threshold, which is critically important in endurance performance.

METHODOLOGY

Participants

Thirty male (age: 21 ± 1.2, body height: 176 ± 4.8 cm, body weight: 69 ± 2.5 kg) physical education and sport students volunteered to participate in this study. All participants were informed about the procedures and signed consent forms before the study. Participants had the same training intensity, they did not have any disease, and they were non-smokers. Participants did not receive any vitamin E preparations in the last 6 months before the study.

Procedures

To examine the effects of 2 weeks vitamin E supplementation to the point of anaerobic threshold, all measurements of the participants were made under physiology laboratory conditions. Participants attended two test periods: pre-test and post-test. Post-test was performed 2 weeks after the supplementation. The experimental methodology is given in Figure 1.

Anaerobic Threshold Determination

Anaerobic threshold of the participants were determined by Conconi method. Conconi et al. defined the relationship between the heart rate and running speed during increasing exercise intensity. The relationship between running speed and heart rate is linear from low intensity to submaximal; however, this linearity deviates when the intensity reaches from submaximal to maximal. At this point, the sudden increase of blood lactate concentration is associated with anaerobic threshold (Conconi et al. 1982).

Pre-test Application

Participants completed 6 minutes of jogging (2 min. 7 km/h speed, 2 min. 7.5 km/h speed, 2 min. 8 km/h speed) on a treadmill (True 750 S.O.F.T. SYSTEM, True Fitness Technology, USA) for warm-up. The subjects' heart rates were recorded during the test procedure with the recording device (Polar Sport Tester Heart Rate Monitor, Polar Vantage Nv, Polar Electro Oy, Finland). After warm-up, participants started the test with initial speed of 8 km/h and running speed was increased by 0.5 km/h for every 200 meters. After every 200 meters, the participants' heart rates were recorded. The test was terminated by researchers when a participant was unable to run. After the test, subjects' anaerobic threshold points were detected using the heart rate and running speed information. After the 24-hour rest period, the same test procedure was repeated. During the second test, when the participants reached their individual anaerobic threshold running speed, the test was stopped and blood was taken from their fingertips and analyzed to detect blood lactate values using blood lactate analyzer (Accutrend Lactate Analyzer, Roche Diagnostics Gmbh, Germany). Blood lactate values on the anaerobic threshold of the participants were recorded.

Post-test Application

After the measurements, the participants were divided randomly into three groups: vitamin E, placebo and control group. The participants who were in the vitamin E and placebo groups took either a vitamin E (single dose vitamin E tablet, containing 400 mg alpha-tocopherol acetate) or a placebo (placebo capsule containing substances in the form of starch) once daily for two weeks. The control group did not receive any substance. At the end of 2 weeks, all the participants in the 3 groups applied again to the pre-test procedure and their anaerobic threshold points and blood lactate values on the anaerobic threshold were determined.

Data are presented as mean ± standard deviation (SD). The differences between pre-test and post-test data were compared using the Wilcoxon test. Kruskal-Wallis test was used to assess the difference between groups and within groups. The Statistical Package for Social Sciences (SPSS 10.0 for Windows) was used for statistical analyses. Statistical significance was set at $p<0.05$.

RESULTS

Descriptive statistics of the participants are presented in Table 1. The average ages of the Vitamin E, Placebo and Control groups was 20.7±1.1 (years), 20.5 ± 0.8 (years), and 21.8 ± 1.4 (years) respectively. The average height of the Vitamin E, Placebo and Control groups was 175.4±4.6 (cm), 179.0±5.2 (cm), 174.0±3.3 (cm) respectively. The average body weight of the Vitamin E, Placebo and control groups were 69.2±2.7 (kg), 70.3±2.7 (kg), and 69.4±2.2 (kg), respectively.

As can be seen in Table 2, results of Kruskal-Wallis test showed that there were no significant differences in the pre-test values of variables between vitamin E, placebo and control groups. When compared to the pre-test values of the ATHR, it seems there was no difference

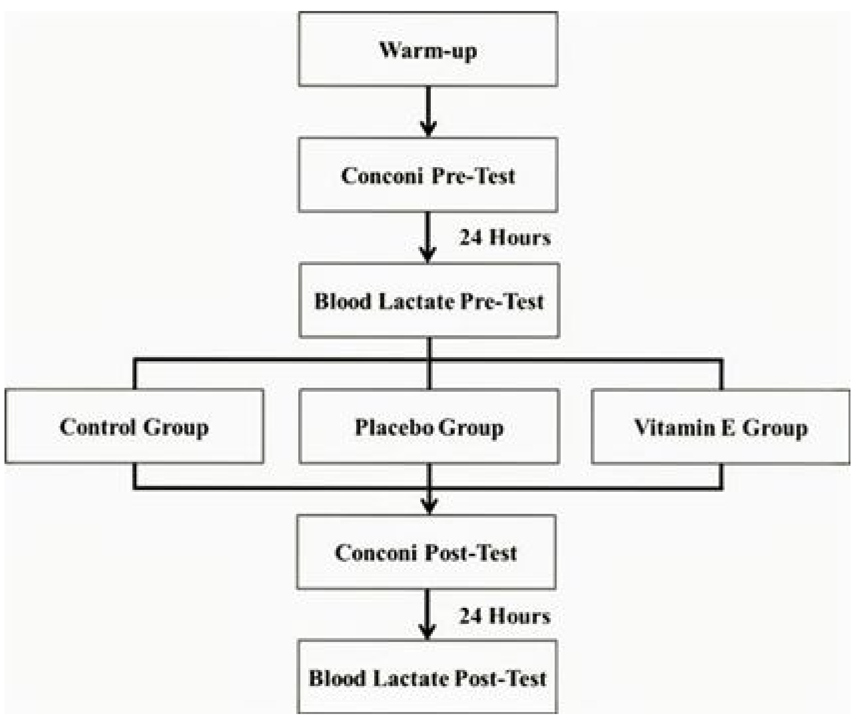

Fig. 1. Experimental methodology
Source: Author

between the groups. The ATHR values of Vitamin E, Placebo and control groups were 179.7±3.9 (beats/min), 177.8±5.1 (beats/min), and 178.6±3.9 (beats/min), respectively. Similarly, there were no significant differences in the pre-test values of ATRS between the groups. The ATRS values of Vitamin E, Placebo and control groups were 12.5±1.3 (km/h), 12.3±0.7 (km/h), and 12.8±1.0 (km/h), respectively. The ATBL values of Vitamin E, Placebo and control groups were 3.3±1.5 (mmol/ml), 4.8±0.9 (mmol/ml), and 4.4±0.9 (mmol/ml), respectively. Pre-test ATBL values were not significantly different between the groups. Compared to the pre-test values of the RDM, it seemed like there were no differences between groups. The RDM values of Vitamin E, Placebo and control groups were 3500.0±335.4 (m), 3340.0±327.2 (m), and 3610.0±425.4 (m), respectively. The HRM values of Vitamin E, Placebo and control groups were 196.4±4.5 (beats/min), 198.7±3.5 (beats/min), and 196.3±3.1 (beats/min), respectively. Pre-test HRM values were similar in all groups (Table 2).

Table 3 presents the pre-test and post-test data comparisons of all study groups. Wilcoxon test results indicate that there was no significant difference between the pre-test and post-test values of variables within the groups. Vitamin E group showed no significant differences between pre and post-test measurements of ATHR, ATRS, ATBL, RDM, HRM. The athr values of Vitamin E group were declined from 179.7±3.9 to 175.7±6.4 (2.2%) after supplementation period, but this decline was not statistically significant (p>.05). Similar results were observed in the ATHR values of placebo; 177.8±5.1 to 177.1±5.1 (0.3%) and control 178±3.9 to 178.4±3.9 (0.1%) groups. According to the pre-test and post-test data comparisons of ATRS, ATBL, RDM, HRM values of all the study groups, there was no significant change observed in any variables. These results indicated that the daily 400mg Vitamin E supplementation for two weeks did not affect the participants' aerobic capacity and blood lactate levels (Table 3).

DISCUSSION

Vitamin E is important for the health of skeletal muscles. Increased oxygen consumption during aerobic exercises causes the formation of free oxygen radicals. Even low intensity strength exercises begin lipid peroxidation (Cakir-Atabek et al. 2015). These free radicals causes damage to the muscle cell's membrane. Vitamin E helps to prevent this damage because of its antioxidant properties (Gerster 1991; Zhang et al. 1998; Vina et al. 2000; Mondal and Bandopadhyay 2016). In the present study, the effects of 2 weeks of vitamin E supplementation (400mg/day) to the point of anaerobic threshold and some of its variables (heart rate at anaerobic threshold, blood lactate at anaerobic threshold, maximal running distance, running speed at anaerobic threshold, maximal heart rate) were examined.

Table 1: Descriptive characteristics of participants

Groups	*n*	*Age (years) (mean ± SD)*	*Height (cm) (mean ± SD)*	*Body Weight (kg)(mean ± SD)*
Vitamin E	10	20.7 ± 1.1	175.4 ± 4.6	69.2 ± 2.7
Placebo	10	20.5 ± 0.8	179.0 ± 5.2	70.3 ± 2.7
Control	10	21.8 ± 1.4	174.0 ± 3.3	69.4 ± 2.2
Total	30	21.0 ± 1.3	176.0 ± 4.8	69.6 ± 2.5

Table 2: Comparison of pre-test values of the participants (Kruskal-Wallis test)

Variables	*Vitamin E(n=10)*	*Placebo(n=10)*	*Control(n=10)*	*p*
ATHR (beats/min)	179.7 ± 3.9	177.8 ± 5.1	178.6 ± 3.9	.62
ATRS (km/h)	12.5 ± 1.3	12.3 ± 0.7	12.8 ± 1.0	.52
ATBL (mmol/ml)	5.3 ± 1.5	4.8 ± 0.9	4.4 ± 0.9	.36
RDM (m)	3500.0 ± 335.4	3340.0 ± 327.2	3610.0 ± 425.4	.24
HRM (beats/min)	196.4 ± 4.5	198.7 ± 3.5	196.3 ± 3.1	.23

p > 0.05

The results of the present study showed no significant change in the observed variables. Two weeks of vitamin E supplementation did not have a positive impact on the participants' anaerobic threshold points. According to the results of the present study, at the end of 2 weeks supplementation period, ATHR values of vitamin E, Placebo, and Control groups were reduced by 2.2 percent, 0.3 percent, 0.1 percent respectively. However, these results were not statistically significant. Many studies prove these findings. According to a meta-analysis by Stepanyan et al. (2014) vitamin E supplementation does not prevent exercise-induced lipid peroxidation and muscle damage clearly. The reason is because of the complex antioxidant structure of tocopherol and its less accumulation in muscle tissue. Dawson et al. (2002) examined the effect of vitamin C and E supplementation on biochemical and ultrastructural indices of muscle damage after a 21 km run. 15 experienced male distance runners were assigned to a vitamin or a placebo group. In the first phase of the study, the vitamin group took 500mg vitamin C and 500 mg vitamin E a day for four weeks. After supplementation period, blood samples were taken of the subjects after 21 km running. In the second phase of the study, vitamin group took 1000mg vitamin C and 1000 mg vitamin E a day for four weeks and the same running period was repeated. As a result, Vitamin E and Vitamin C supplementation had no role in any reduction in the level of biochemical and cellular enzymes that cause muscle damage after a half period of marathon in experienced male runners. Itoh et al. (2000) conducted a study on fourteen male runners who were supplied with vitamin E (1200IU/day) or placebo. At the end of 6 days maximal treadmill training, they did not find a significant difference in maximal oxygen uptake (VO2 max), and maximal heart beats between vitamin E and placebo groups. Rokitz et al. (1994) conducted a study on 30 senior bike riders using the ergometer; they found no significant changes in heart rates of the subjects with vitamin E. In one study Morrison et al. (2015) conducted in eleven healthy young males to determine the effects of vitamin C and vitamin E supplementation on endurance performance and oxidative stress of skeletal muscle, the results suggest that following acute exercise, supplementation of vitamin C (2x500 mg/day) and vitamin E (400 IU/day) did not attenuate skeletal muscle oxidative stress. In another research, Mohammed et al. (2015) investigated the effects of dietary vitamin C and E supplementation on oxidative stress and muscle damage markers among male and female weight lifters. They designed the study to be randomized, single blind, and placebo controlled. After 6 weeks of weightlifting training intervention, blood and urine samples were tested. According to the findings of the study, antioxidant supplementation did not provide any benefit to reduce muscle damage and oxidative stress.

In contrast, some other investigations showed the beneficial impact of antioxidants supplementation on muscle damage and oxidative stress. In one of them, Santos et al. (2016) evaluated the effects of vitamin E supplementation on muscle damage and inflammation after

Table 3: Comparison of pre-test and post-test values of the participants. (Wilcoxon signed-rank test)

Variables	*Vitamin E*			*Placebo*			*Control*		
	Pre	*Post*	*p*	*Pre*	*Post*	*p*	*Pre*	*Post*	*p*
athr (beats/min)	179.7±3.9	175.7±6.4	.06	177.8±5.1	177.1±5.1	.22	178.6±3.9	178.4±3.9	.41
atrs (km/h)	12.5±1.3	12.6±1.3	.31	12.3± 0.7	12.3±0.8	.70	12.8±1.0	12.7±1.0	.41
atbl (mmol/ml)	5.3±1.5	5.5±1.4	.87	4.8±0.9	4.7±0.8	.57	4.4±0.9	4.5±0.9	.30
rdm (m)	3500± 335	3580± 614	.15	3340±327	3380±319	.31	3610±425	3580±436	.43
hrm (beats/min)	196.4±4.5	196.3± 4.1	.78	198.7±3.5	199.2±2.8	.18	196.3±3.1	196.4±2.8	.65

exercise in hypoxia. Nine participants took vitamin E (250 mg) before 60 minutes of exercise under hypoxic condition for one week. At the end of the exercise period, the results indicated that 250 mg vitamin E supplementation reduces cell damage markers in hypoxia. In a study by Park and Kwak (2016), they reported that both aerobic and anaerobic exercise training are beneficial to improve redox balance in humans, and any type of exercise training will be beneficial for improving antioxidant defense system in athletes. Mondal and Bandopadhyay (2016) investigated the effect of the combination of vitamin E and exercise on some hematological parameters. Ninety six university male students participated in the study and were randomly assigned to control, treadmill exercise, treadmill exercise and Vitamin E groups. After ten weeks intervention period, blood samples were taken from the subjects. The results revealed that, combined Vitamin E and treadmill exercise supplementation altered the hemoglobin content and this can positively affect exercise performance. In another study, Huebsch et al. (1997) found that the daily use of 335 mg of vitamin E lowered blood pressure during aerobic exercise.

According to data obtained from the present study, after 2 weeks supplementation period, ATBL values of participants were not significant. The differences were 1.8 percent increase in the vitamin E group, 1.8 percent decrease in the placebo group and no changes in the control group. There are some studies in literature that have similar findings along with the present study. Lawrence et al. (1975) conducted a study on swimmers. Subjects used 900 IU alpha-tocopherol (vitamin E) and placebo daily. After swimming endurance test, they found no significant difference between serum lactic acid levels of subjects. In the study of Rokitz et al. (1994), similar results are seen. There was no significant difference between vitamin E and placebo groups on the ATBL. In another research, Surmen et al. (1999) studied the effect of vitamin E supplementation on post-exercise plasma lipid peroxidation and blood antioxidant status in smokers. In this study, subjects were given 400 mg vitamin E daily for 28 days. After maximal exercise test, the plasma level of lactate were not significant between the vitamin and placebo groups. In the study by Silva et al. (2010), twenty-one participants were divided randomly into placebo and supplemented groups. After supplementation period, (800 IU vitamin E for daily 3 weeks) the participants performed eccentric exercise (elbow flexion-extension) until exhaustion. According to the results of blood sample, vitamin E supplementation showed a significant effect against muscle damage and oxidative stress.

Lipid peroxidation starts when free radicals increase during exercise. In this case, vitamin E requirement increases for preventing oxidative damage. Vitamin E can delay muscle fatigue by preventing free radical damage and may affect performance positively (Dincer and Kayserilioglu 1995). The results of the present study showed no significant changes in the RDM and ATRS. Maximal running distance (RDM): 2.2 percent increase for vitamin E, 1.1 percent increases for placebo, and 0.8 percent decreases for control. Running speed at anaerobic threshold (ATRS): 0.8 percent increase for vitamin E, 0.7 percent decrease for control, and no change in placebo. In contrast to our study, Hoque and Bandopadhyay (2016) conducted a study in thirty male 19-24 year old 100m and 200m sprinters. The subjects used vitamin E 5 times per week for 6 weeks. After supplementation period, the subjects performed running based anaerobic sprint test. According to the results of study, Vitamin E supplementation developed anaerobic power and gave the sprinters a better sprinting ability.

According to another data obtained from the present study, vitamin E did not affect the subject's maximum heart rate during the Conconi test. HRM 0.5 percent decrease for vitamin E, 0.2 percent increase for placebo, and 0.05 percent increase for control. These changes were not significant. Generally, the maximum heart rate is unchanged. As a result of endurance training; resting and submaximal heart rate is reduced significantly. Endurance training does not change maximum heart rate. Even maximal heart rate may drop slightly after endurance training (Sonmez 2002). The results of this study indicate that maximum heart rate is age dependent and training effect is minimal.

CONCLUSION

According to the findings of the present study, use of 400 mg vitamin E daily for two weeks has no positive effects on the endurance performance of the participants. Anaerobic threshold and blood lactate values of the anaerobic threshold was not seen as a positive development.

RECOMMENDATIONS

Some limitations in the current study included conducting the study only on young males and the amount of vitamin supplementation was limited to 2 weeks 400mg/day. Future research within young females and different age groups may reveal different results. In addition, long-term effects of vitamin E supplementation beyond 2 weeks may be considered.

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