

The Effect of Vitamins C and E Supplementation on Muscle Damage, Performance, and Body Composition in Athlete Women: A Clinical Trial

Maryam Taghiyar^{1,2}, Reza Ghiasvand^{1,2}, Gholamreza Askari^{1,2}, Awat Feizi³, Mitra Hariri^{1,2}, Nafiseh Shokri Mashhadi⁴, Leila Darvishi^{1,2}

¹Food Security Research Center, Isfahan University of Medical Sciences, Isfahan, Iran, ²Department of Community Nutrition, School of Nutrition and Food Science, Isfahan University of Medical Sciences, Isfahan, Iran, ³Department of Biostatistics and Epidemiology, School of Health, Isfahan University of Medical Sciences, Isfahan, Iran, ⁴Child Growth and Development Research Center, Isfahan University of Medical Sciences, Isfahan, Iran,

Correspondence to:

Mrs. Darvishi Leila,
Department of Nutrition, School of Food Science and Nutrition, Isfahan University of Medical Sciences, Isfahan, Iran.
E-mail: leilad_78@yahoo.com

Date of Submission: Mar 10, 2012

Date of Acceptance: Jun 15, 2012

How to cite this article: Taghiyar M, Ghiasvand R, Askari G, Feizi A, Hariri M, Mashhadi NS, *et al.* The effect of vitamins C and E supplementation on muscle damage, performance, and body composition in athlete women: A clinical trial. *Int J Prev Med* 2013; Supplement 1: S24-30.

ABSTRACT

Background: Due to the special training conditions and lifestyle athletes require an antioxidant system that is more efficient than others. To keep this system optimal, many of them use antioxidant supplements. This study aimed to investigate the effects of vitamins C and E supplementation on muscle damage, performance, and body composition in athlete women.

Methods: The study was a 4-week randomized, double-blind clinical trial conducted on 64 trained female athletes recruited in Isfahan sports club. They were randomly assigned to one of the following four groups; A: vitamin C (250 mg/day), B: vitamin E (400 IU), C: vitamin C + vitamin E and control (placebo). Harvard Step Test was used to measure maximal oxygen consumption for performance, body composition, and damage marker (myoglobin) were measured before and after the intervention.

Results: Comparing the result of the test in performance of sport, there was no significant difference between groups in VO_2 max. Also, vitamin supplements had no significant effect on subcutaneous fat between the groups, however, in the intergroup comparison, were significantly increased in group control ($P = 0.03$). But, there were no significant differences, change in myoglobin between the groups. There was a significant increase in group A ($P = 0.04$).

Conclusions: Vitamins C and E supplementation had no significant effect on any of the studied parameters.

Keywords: Body composition, clinical trial, vitamin supplementation, female athlete, muscle damage, performance, vitamin C, vitamin E

INTRODUCTION

Resistance training is reported to have many advantages such as weight control, prevention of osteoporosis, improvement of cardiovascular risk factors, and prevention of injury.^[1,2]

However, excessive resistance training programme may increase oxidative stress and cellular damage.^[3] Free radicals, which are atoms and molecules that have an unpaired electron, can damage molecules that are important for cellular function, leading to a total loss of cellular function.^[4] Free radicals attack the cells of critical ingredients such as lipids, proteins, and

DNA.^[5] Results in the formation of superoxide radicals, 1 to 3 percent oxygen is converted to reactive hydroxyl radicals. The result, oxygen consumption increased electron transport through the respiratory chain and ultimately increases the production of free radicals.^[6] Antioxidant systems of the human body through the production and use of antioxidants, disconnect the chain reaction caused by free radicals.^[7] The antioxidants react with free radicals; free radicals are converted weak and have no desire to react with surrounding molecules.

Endurance athletes due to special circumstances, require an antioxidant system efficient than others; the system maintains normal function of the body and adjust to increased oxidative stress induced by free radicals.^[8] There are two ways of antioxidant defense. One, physiological endogenous antioxidant systems, including glutathione peroxidase, catalase, and superoxide dismutase enzymes,^[7,9] and another defense against oxidative damage, antioxidants are derived from food sources.

It is very important to have a proper diet for athletes to improve athletic performance of a certain antioxidant systems after intense exercise, although athletes may be unable to provide adequate antioxidant substances needed by the body from the diet. Therefore, antioxidant supplements can be used for such individuals. The best recognized ones are vitamins C and E antioxidants, among that are used mainly for sports supplements.^[10] Vitamin C effectively reduces free radicals and also helps to restore the vitamin E. This vitamin also helps with iron absorption and has an important role in the formation of creatine, which is the main source of muscle energy. Vitamins E and C have a role in strengthening and increasing with endogenous antioxidant glutathione and lipoic acid; these two substances have a major role in the reconstruction of important vitamins from their radical forms.^[4]

Some studies have been conducted on exercise performance using antioxidant supplements known to improve performance^[11] But some of them don't show any effect of antioxidant supplements on athletic performance and even expressed delayed recovery damaged muscle with the supplements, but antioxidant supplement still may be effective on exercise performance.^[12] Further research is needed to answer whether supplements are helpful for athletes,^[13] especially female athletes, and limited

studies have been done in this field. Therefore, this study is aimed to investigate the effects of vitamin C and E supplementation on muscle damage, performance, and body composition in athlete women.

METHODS

Sixty-four trained (the best or most skilled members who had been doing aerobics for at least 3 years) female athletes with the inclusion criteria were randomly recruited in Isfahan sports club. The study was approved by the Ethics Committee of Isfahan University of Medical Sciences. This study was registered in the Iranian Registry of Clinical Trials (www.irct.ir) on October 7, 2012 with IRCT registration number: IRCT201111095062N2. Considering MDA as the main variable in the study sample and at the significance level of 5% and statistical power of 80%, to detect a measurable amount of 1.2, 12 subjects were assigned to each of the groups. Subjects were randomized using permuted of size 4 blocks by another person to one of four groups; A: vitamin C (250 mg/day), B: vitamin E (400 IU), C: vitamin C + vitamin E, and control (placebo). They were initially willing to participate in the study and already had 3 to 6 years of experience in sports, were not taking antioxidant supplements in the previous month, were in complete physical health, and were free of disorders related to pregnancy and lactation. During the study, one participant was excluded from the sample due to a fracture in the leg. Moreover, one participant due to digestive problems and weight changes and four participants withdrew from groups due to personal reasons. After explaining the purpose and details of the sample, written informed consent was obtained from each subject. Each participant's weight and height were measured and recorded using medical scales Seca (Made in Germany) with an accuracy of 100 grams and stadiometer with an accuracy of 0.5 cm.

The study was a 4-week randomized, double-blind clinical trial. Athletes consumed a vitamin pill daily with their meals; vitamin C (Iran, Osveh) or vitamin E (Iran, Zahravi). Vitamins placebo were prepared by the same company. In the beginning, each participant took supplements for four weeks and by counting the

total number of pills remaining in the study, it was figured out if the participants followed the research code or not consumed. They were asked to keep doing their daily routine physical activity without changing it using a two-day food record. The information about their diet was collected at the outset and in the last week of intervention and analyzed with nutritionist software, version 4. Physical activity was assessed using a physical activity questionnaire and described according to metabolic equivalent-minutes a week standard. Fat were also measured by skinfold caliper (Johnson, England) at four points; triceps, biceps, supra iliac, and subscapular.^[14] On the other hand, VO_2 max was measured via Harvard step test.^[15]

Blood biochemical evaluation was performed at the baseline and at the end of the fourth week before giving supplements to participants. Blood samples of 10 ml were taken from the brachial vein. To separate serum and blood samples, the samples were stored at room temperature for 1 hour and then centrifuged at 2000 rpm for 15 minutes. After that, serum samples were stored at $-70^{\circ}C$ before they were analyzed. Myoglobin was measured by Human Myoglobin (MYO) ELISA Kit (Co.Glory Science). Analyses were performed with SPSS software, version 16 statistical package. The results are presented as mean \pm standard error. Due to the lack of normal distribution of variables, they were log transformed for normalization. Method of multivariate analysis of variance was used to examine differences (percentage changes before and after the intervention) of variables among groups. Paired *t*-test was used to compare changes within groups of variables and Bonferroni test was used to compare changes between groups as a method of post-hoc comparisons of them.

RESULTS

Descriptive information about the sample is shown in Table 1. The age of the participants of the study was significantly different among the groups ($P < 0.05$) but BMI and physical activity were not between groups [Table 1].

Compared the performance of sport, there was no significant difference between groups in VO_2 max. Also, vitamin supplements had no significant effect on subcutaneous fat between the groups, however, in the intergroup comparison, were

Table 1: Descriptive information

| Variable | Groups | Number | Mean \pm SE | P |
|----------------------------------|--------|--------|-------------------|----------|
| Age (years) | 1 | 16 | 31.3 \pm 1.8 | 0.007*** |
| | 2 | 13 | 38.5 \pm 1.6 | |
| | 3 | 14 | 33.9 \pm 1.5 | |
| | 4 | 15 | 38.1 \pm 1.4 | |
| | Total | 58 | 35.3 \pm 0.9 | |
| BMI (kg/m ²) | 1 | 16 | 26.7 \pm 1.1 | 0.8 |
| | 2 | 13 | 25.4 \pm 1.09 | |
| | 3 | 14 | 25.8 \pm 1.2 | |
| | 4 | 15 | 25.7 \pm 0.8 | |
| | Total | 58 | 25.9 \pm 0.5 | |
| Physical activity (Met.min/Week) | 1 | 16 | 636.1 \pm 93.2 | 0.1 |
| | 2 | 13 | 676.1 \pm 137.1 | |
| | 3 | 14 | 997.3 \pm 160.6 | |
| | 4 | 15 | 659.6 \pm 145.9 | |
| | Total | 58 | 736.3 \pm 68.38 | |

*** $P < 0.01$

significantly increased in group control ($P = 0.03$). But, there were no significant differences, change in myoglobin between the groups. But there was a significant increase in group 1 ($P = 0.04$) [Table 2].

Dietary intake was assessed using a two-day food records during Software by Nutritionist 4 that there was no significant difference between groups and within groups [Table 3]. The aim was to compare intake of vitamins A, E, C, D, β -Caroten, niacin, selenium, copper and zinc between the groups. The only significant difference was seen on selenium ($P < 0.05$), which had no effect on the other variables [Table 3]. We show the schematic diagram of this study in [Figure 1].

DISCUSSION

The purpose of this study is to investigate the effects of vitamin C and E supplementation on muscle damage, performance, and body composition in athlete women. In a similar study, Yfanti *et al.* (2010) investigated the effects of combined vitamin C and E supplementation to healthy individuals on different measures of exercise performance after endurance training. Muscle biopsies were obtained before and after training. The results suggested that administration of vitamins C and E to individuals with no previous vitamin deficiencies has no effect on physical adaptations to strenuous endurance training.^[16]

Dawson *et al.* (2002) investigated whether 4 weeks of daily supplementation with 500 or 1000 mg of vitamin C and 500 or 1000 IU of vitamin E could modify biochemical and ultrastructural indices of muscle damage following a 21 km run in 15 experienced males. They revealed that vitamin C and E supplementation (500 or 1000 mg or IU per day) for four weeks does not reduce either biochemical or ultrastructural indices of muscle damage in experienced runners after a half

Table 2: Mean±SE: Indicators of muscle damage, performance, and body composition compared between and within groups

| Groups | Mb (µg/L) | VO _{2max} (ml.kg ⁻¹ .min ⁻¹) | Fat(%) |
|----------------------|-------------|---|------------|
| Group 1 | | | |
| Pre | 81.7±17.9 | 47.2±0.6 | 42.5±0.7 |
| Post | 115.3±3.4 | 47.3±0.5 | 42.5±0.7 |
| changes ¹ | 33.5 | 0.1 | -0.03 |
| P ¹ | 0.04** | 0.9 | 0.8 |
| Group 2 | | | |
| Pre | 60.01±15.8 | 48.04±0.8 | 41.6±1.3 |
| Post | 56.8±8.7 | 47.9±0.58 | 41.6±1.36 |
| changes ¹ | -3.1 | -0.06 | -3.1 |
| P ¹ | 0.5 | 0.9 | 0.9 |
| Group 3 | | | |
| Pre | 159.2±40.05 | 45.9±0.6 | 41.7±0.9 |
| Post | 182.1±4.8 | 45.8±0.7 | 41.9±0.8 |
| changes ¹ | 22.8 | -0.1 | 0.1 |
| P ¹ | 0.2 | 0.8 | 0.2 |
| Group 4 | | | |
| Pre | 74.4±19.9 | 48.36±0.8 | 42.02±1.18 |
| Post | 84.7±30.3 | 48.39±0.6 | 42.3±1.13 |
| changes ¹ | 10.3 | 0.03 | 0.3 |
| P ¹ | 0.4 | 0.9 | 0.03** |
| P value ² | 0.4 | 0.9 | 0.9 |

P¹ inter-group comparisons, P² between-groups comparison, ¹The mean difference between the beginning and end, ***P<0.01, **P<0.05, *P<0.1

Table 3: Mean±SE dietary intake

| Groups | Vit A | Vit E | Vit C | Vit D | β-caroten | Niacin | Se | Cu | Zn |
|---------|--------------|-----------|------------|---------|-------------|-----------|------------|----------|----------|
| Group 1 | 1708.6±850.2 | 12.4±1.2 | 114.4±89.9 | 1.3±1.4 | 373.1±88.9 | 22.9±14.8 | 0.08±0.01 | 1.01±0.4 | 9.01±7.9 |
| Group 1 | 472.8±59.1 | 14.6±2.8 | 88.5±63.2 | 0.8±1.5 | 54.4±14.04 | 17.1±5.9 | 0.04±0.003 | 0.9±0.3 | 6.4±2.6 |
| Group 3 | 789.7±232.2 | 14.1±4.06 | 117.7±62.4 | 1.3±1.5 | 447.7±195.5 | 16.9±3.8 | 0.06±0.008 | 1.07±0.4 | 7.3±1.7 |
| Group 4 | 511.5±137.9 | 13.6±5.8 | 99.3±74.1 | 0.8±2.5 | 263±113.6 | 15.4±6.1 | 0.04±0.007 | 0.8±0.1 | 5.5±1.9 |
| P | 0.7 | 0.6 | 0.6 | 0.7 | 0.1 | 0.3 | ***0.007 | 0.2 | 0.1 |

***P<0.01

marathon.^[17] Weight *et al.* (1998) examined whether a multivitamin and mineral supplement influenced the athletic performance of 30 competitive male athletes. At 0, 3, 6, and 9 mo the runners performed a progressive treadmill test to volitional exhaustion for measurement of maximal oxygen consumption, peak running speed, blood lactate turn point, and peak post exercise blood lactate level. Running time in a 15 km time trial was also measured. They showed that 3 month of multivitamin and mineral supplementation was without any measurable ergogenic effect.^[18] Telford *et al.*^[19] examined the effect of vitamin and mineral supplementation over 7 to 8 months of training and competition in 82 athletes from four sports: basketball, gymnastics, rowing, and swimming. All athletes were monitored to ensure that the recommended daily intakes (RDI) of vitamins and minerals were provided by diet alone. Sport-specific and some common tests of strength as well as aerobic and anaerobic fitness were performed. Coaches' assessment of improvement was also obtained. In general, this study provided little evidence of any effect of supplementation to athletic performance for athletes consuming the dietary RDIs.^[19] Singh *et al.* (1992) investigated the effects on physical performance of 90 d of supplementation with a high potency multivitamin-mineral supplement in 22 healthy, physically active men. They randomly assigned subjects to a supplement or placebo group; both groups had similar physical characteristics. Performance was assessed from maximal aerobic capacity, endurance capacity, and isokinetic tests. The results suggested that supplementation did not affect physical performance in well-nourished men who maintained their physical activity.^[20]

Some researchers have reported different results. Aguiló *et al.* (2007) examined the effects of antioxidant diet supplements on blood lactate concentration and on the aerobic and anaerobic

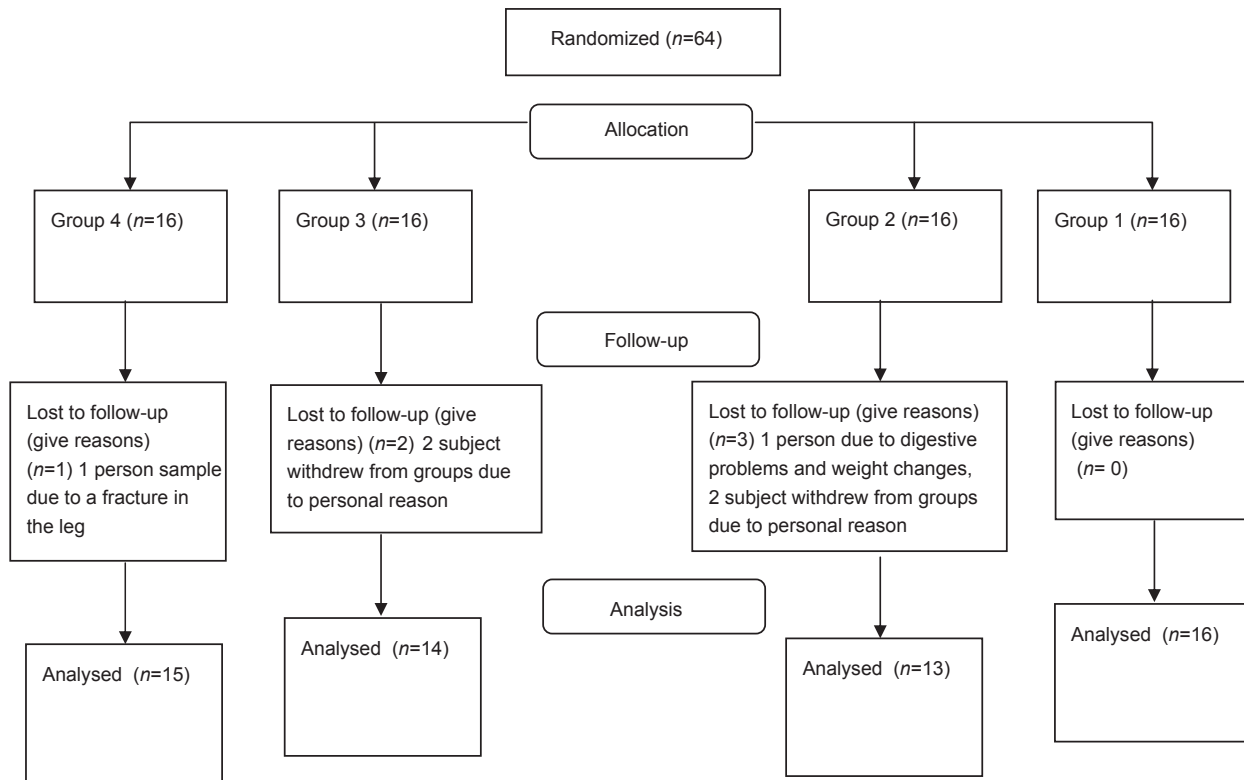


Figure 1: Schematic diagram of the study

thresholds and their adaptations to training were analyzed in 15 amateur male athletes. In the study, before and after the antioxidant supplements, the sportsmen performed a maximal exercise test on a cycle ergometer and maximal and submaximal physiological parameters were assessed together with blood lactate concentration. They revealed that antioxidant diet supplements induced lower increases in blood lactate concentration after a maximal exercise test and could improve the efficiency in which aerobic energy is obtained.^[21]

MacRae *et al.* (2006) investigated whether 6 wk of antioxidant supplementation (AS) would enhance 30 km time trial (TT) cycling performance in eleven elite male cyclists. The study to test the effects of twice daily AS containing essential vitamins plus quercetin (FRS) and AS minus quercetin (FRS-Q) versus a baseline TT (B). Thus, FRS supplementation significantly improved high-intensity cycling TT performance through enhancement of power output.^[22] Kon *et al.* (2008) examined the effects of an antioxidant, coenzyme Q10(CoQ10), on muscular injury and oxidative stress during exercise training in 18 male students. They showed that indicate that CoQ10 supplementation

reduced exercise-induced muscular injury in athletes.^[23] Yu *et al.* (2003) investigated the effect of antioxidant vitamins (vitamin E and vitamin C) on the exercise performance of rats. Fifty male SD rats were randomly divided into control group (C), exhausting exercise control group (E), vitamin E group (M1), vitamin C group (M2), and vitamin E plus vitamin C group (M3). The rats in the exercising groups (E, M1, M2, M3) were propelled for repeated exhausting runs on the treadmill for 4 weeks.

They revealed that vitamin E can protect the mitochondria in the skeletal muscles and improve the exercise performance of rats, the effect of which can be enhanced by vitamin C, but vitamin C alone cannot sufficiently achieve the effects.^[24,25,26] In the present study, there were no significant changes in the performance of athletes; this could be due to different types of exercise and antioxidant supplementation or supplementation time or gender difference. The majority of studies in this area have been carried out on male athletes than female. Also, a study conducted in China, and the result was an improved performance consumption of vitamin supplements, and a design very

similar to this study, has been done on laboratory animals. This study could also be designed to examine the separate vitamins E, C and combined model of these vitamins and with the same study population, female athletes. However, there was a limit in the number of samples in this study, since more samples would yield more reliable results so this point is suggested to be considered in further related researches. Future studies are suggested to be conducted with larger sample size and on other athletes using different antioxidant supplements.

CONCLUSION

Generally speaking, the results of this study showed that supplementation with vitamin C and E supplementation in groups had no significant effect on any of the parameters studied.

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Source of Support: This study was conducted as a thesis funded by Isfahan University of Medical Sciences, Isfahan, Iran,
Conflict of Interest: None declared.

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