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The effect of four weeks vitamin C supplementation with intermittent exercise on serum anti-oxidation capacity and maximal oxygen consumption in inactive women

Mandana Gholami, Negin Arab* and Farshad Ghazalian

Department of Physical Education and Sport Sciences, Science and Research Branch, Islamic Azad University, Tehran, Iran .
Corresponding Author: Negin Arab, M.Sc

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ABSTRACT

Present study carried out to determine the effect of four weeks vitamin C supplementation with intermittent exercise on serum anti-oxidation capacity and maximal oxygen consumption in inactive women. 40 healthy inactive women after the aerobic capacity detection were randomly set in four equal groups of recipients of supplemental vitamin C (1000 mg daily for two meals a day for four weeks) (CG), placebo (lactose) (PG), interval training with placebo (three days a week, with 70% of maximum heart rate, received a five point nine minutes, four minutes passive rest) (P+TG) and interval training with vitamin C (C+TG). Initial blood sample obtained at baseline before starting supplementation and second blood sample was taken after completion of supplementation and intermittent exercise. Normal data were investigated by using one-way analysis of variance and post Bonferroni test, if significant, T in the five percent level of significance using SPSS version 22. **Results:** Four weeks of vitamin C supplementation on serum anti-oxidation capacity of inactive women has a significant effect (P = 0.04). But has no significant effect on maximal oxygen consumption (P = 0.11). The four-week interval training has a significant effect on serum anti-oxidation capacity (P = 0.001) and oxygen consumption (P = 0.001) on maximum inactive women. Four weeks vitamin C supplementation with intermittent exercise on serum anti-oxidation capacity (P = 0.001) and oxygen consumption (P = 0.0001) has a significant effect on inactive women. **Conclusion:** The results of this study also suggest that three days in a week, with 70% of maximum heart rate, received five nine minutes session, with four minutes inactive rest significantly increased serum anti-oxidation capacity and maximal oxygen is consumed. In addition, the four-week vitamin C supplementation, with promotion of serum anti-oxidation play an important role in the prevention of adverse changes in oxidative stress, membrane damage and loss in peak muscle damage after eccentric activity.

Keywords: *Vitamin C, Total anti-oxidation Capacity, Interval training, maximal oxygen consumption, inactive women.*

INTRODUCTION

Considering the beneficial effects of physical activity on quality of life and increased production of free radicals from these interactions, it is likely that the free radicals production due to physical activities have essential intervention in people health. Research results suggest that high consumption of oxygen during exercise, increase the production of oxygen species (ROD). Increased in production of free radicals during irregular and prolonged exercise leads to fatigue, inflammation and tissue damage (19). In addition, many studies show that Increased oxidative stress in the body and damage caused by oxidation reactions of nucleic acids, proteins, enzymes and other molecules could lead to the development and exacerbation of degenerative diseases such as cardiovascular disease, diabetes, cancer, Alzheimer's, Parkinson's, cataract and aging (13). Also, scientific evidence has shown that oxidative stress caused by prolonged exercise causes tissue damage and initiate inflammatory responses (8). Hence, most scientists in particular life sciences researchers plan to assist athletes and promote human development by identify the various sports and offering practical strategies. One way to prevent and reduce oxidative stress and its consequences is using the anti-oxidation additives. But most research has been done in the field on anti-oxidation additives industry. On the other hand, some studies have shown that industrial supplements long-term usage have defects for the body (19). For this reason, today, in order to maintain and enhance the health of consumers as well as access to new resources and cheap natural antioxidant, research

in this area is necessary. Many studies have reported that vitamin C has anti-oxidation compounds. However, long-term use of vitamin C can boost muscle tissue collagen (2). On the other hand, Bloomer et al research (2006) noted that taking 1,000 milligrams of vitamin C per day for two weeks, statistically hasn't significant changes in Malondialdehyde after two and a half hours of cycling at 60% of maximum oxygen consumption (5). However, the effective dose of vitamin C for four weeks period and to the impact of periodic research activity there is no thing for record and it is essential to fill this space. Thus, according to the need of improving the daily life and to make better use of the benefits of physical activity by non-athletes and athletes and reduce the symptoms of exercise-induced oxidative stress, where necessary, to a four-week vitamin C supplementation on total anti-oxidation capacity and maximum oxygen consumption of inactive women be determined in periodic training.

Materials and Methods

Present study conduct in semi-experimental design in four groups (CG, PG, P+TG, C+TG) with two stages measurement in double blind type. The study population included healthy women, non-athletes (no regular participation in physical activity and exercise during the last six months) and non-smokers in Tehran with an average age of 25 ± 3 years. After distributing leaflets and cooperation between women's participation in the research project in the city of Tehran, 100 volunteers announced that 40 candidates were qualified to participate in the research project. Volunteers attended in the coordinating meeting and after explain the objectives and methods of measuring by the researcher, with the informed consent form and health questionnaire and 24 hour dietary recalls, Medical examinations were performed. Two weeks before the start of the study, the anthopometric indicators (body measurement), height, weight and percentage of body fat using skin calipers thickness gauge of Japanese Mikusha (Skinfold Calipers) and three-point formula of American College of Sports Medicine (above on the right arm triceps, abdominal and pelvic skin folds) was measured. After determining the thickness of the skin pin, three times the average size of each part of the body was placed in the following formula.

Fat percentage = (0.39287) * (sum of three parts) – 0.00105 * 2 (sum of three parts) + [0.15772*(age)]-5.18845

The maximum oxygen consumption by the Cooper test (Bruce) on treadmill (Made in Italy brand TechnoJim) was determined using the following formula.

Maximum oxygen consumption = (mileage m – 504.9) / 44.73

Then, randomly divided into two equal groups receive supplements of vitamin C (1000 mg effervescent tablets of 500 mg twice daily) and placebo were replaced (17). Vitamin C products was prepared from Osveh Pharmaceutical Co. Tehran with product license number 11.10103. Also the 24-hour dietary survey questionnaire was used to remind vitamin C consumption. Initial blood samples collected at baseline before starting supplementation from right arm elbow vein, all subjects were taken at the 8-9 a.m. in Laleh hospital. The exercise protocol (three days a week, with 70% of maximum heart rate, each session include five nine minutes level, four minutes of inactive rest) for four weeks, starting at the last session of the second blood sample was taken in the fourth week.

Sampling method:

Blood in each of the three subjects was taken 3 milliliter of blood in sitting position from a right elbow vein and was poured in test tubes without anticoagulant substance. For the serum preparation, samples were incubated for 10 min at room temperature and then serum was separated from clot by centrifugation in 3000 rpm for 10 minutes. Test samples to the total anti-oxidation capacity (TAC) were kept in the freezer -20 ° C. All stages were performed in same condition (8 to 9 am, the temperature of 28-26 ° C and humidity 50%). In addition, participants 48 hours before the test avoid from do any heavy physical activity and meals were the same before the test.

Measurement of serum total anti-oxidation capacity using Randox kits with MX2332 product number by the auto analyzer (America) at a wavelength of 600 nm was performed.

Statistical analysis:

At first the normal data distribution evaluated by Kolmogorov-Smirnov test; since the data distribution was normal, describing data was expressed average and standard deviation. Then change any of the parameters analyzed during different stages of measurements using one way ANOVA post hoc Bonferroni test and, if significant, independent t-test were analyzed. Statistical analysis was perform at five percent significant level using SPSS statistical software version 21 and Excel 2013.

RESULTS AND DISCUSSION

Results

The questionnaire was distributed to 100 volunteers were ready to participate in the study that conditions of participation was 40, for each group, 10 patients were allocated to individual characteristics. (Table 1)

Table 1. Mean ± SD of physiological and anthropometric characteristics of the subjects

Indicators / Groups	Age (years)	Weight (Kg)	Height (cm)	Body mass index (k.m^2).	Fat (%)	Waist-hip (cm)
CG	22.4 ± 1.17	70.5 ± 3.2	160.5 ± 3.68	23.29 ± 2.9	22.72 ± 2.5	0.87 ± 0.04
PG	23.4 ± 1.64	73.1 ± 8.39	161± 3.3	24.28 ± 2.95	23.4 ± 2.07	0.83 ± 0.04
P+TG	24.4 ± 1.71	74.4 ± 7.39	161.4 ± 2.36	22.43 ± 2.09	23.4 ± 1.07	0.84 ± 0.04
C+TG	23.5 ± 1.15	71.5 ± 4.22	161.1 ± 2.42	24.26 ± 2.9	22.72± 3.66	0.86 ± 0.04

The age distribution in both groups were close, in terms of body mass index and body fat percentage were normal.

Table 2. Check ANOVA for indicators

Indicators measure		Sum of Squares	df	Mean Square	f	Sig.
total anti-oxidation capacity	Between Groups	3.15	3	1.1		
	Within Groups	3.37	36	0.09	11.77**	0.0001
	Total	6.69	39			
Vo_{2max}	Between Groups	355.22	3	118.4		
	Within Groups	284.09	36	8.6	13.75**	0.0001
	Total	639.32	39			

** Indicates significant at 1% level

According to Table 2 ANOVA measurement for both indexes was significant, therefore, to better understand the significant differences between any groups we use post hoc Bonferroni test that according to Table 3, it is observed that there was a significant difference between the total anti-oxidative capacity for the group "placebo with exercise and placebo" "placebo with exercise and supplements", " supplements with exercise and placebo," "Supplementation with practice supplement " and as well as maximum oxygen consumption index between "like drug and placebo exercise", "exercise and supplements with placebo," "supplement and placebo exercise", "Supplementation with exercise and supplements" have significant differences.

Table 3. Bonferroni post hoc test

Indicators measure	Groups	Groups	Mean Difference	Std. Error	Sig.
total anti-oxidation capacity	PG	CG	0.11	0.12	1
		P+TG	0.58*	0.13	0.001
		C+TG	0.66**	0.13	0.0001
	CG	P+TG	0.46*	0.13	0.009
		C+TG	0.54*	0.13	0.002
	P+TG	C+TG	0.07	0.13	1
Vo_{2max}	PG	CG	0.16	1.44	1
		P+TG	4.59*	1.44	0.01
		C+TG	7.43**	1.44	0.0001
	CG	P+TG	4.43	1.31	0.01
		C+TG	7.18**	1.31	0.0001
	P+TG	C+TG	2.75	1.31	0.26

* Indicates significant at 5% level

** Indicates significant at 1% level

Discussion

This study shows that there is a difference between four groups of serum anti-oxidation capacity of inactive women. Total anti-oxidation capacity changes in the placebo group and placebo with training, exercise and supplements also been significant. The Total amount of anti-oxidation capacity in the exercise and supplements group is more than the other groups. Because it can stimulate the supplement, as well as training stimulates anti-oxidation total capacity. In the meantime, the team practiced supplement and placebo groups, as well as exercise and supplements are also significant. Therefore, we can conclude that the group exercise and supplements have the highest increase compared to other groups. This study also showed that four weeks vitamin C supplementation significantly increase total anti-oxidation capacity. among researches that perform in this context which match with this studies results Oberbach et al. (2010), Wadley et al. (2010), Rosa et al (2009), Kelkar et al (2008) could be noted (23,22,11,20). In Oberbach et al research, 1,000 milligrams daily intake of vitamin C and 400 international units of vitamin E within four weeks cause an increase in anti-oxidation capacity at the basic state (20). The mechanism proposed in relation to the effects of vitamin C supplementation on serum total anti-oxidation capacity in this case, vitamin C directly interact

with superoxide and hydroxyl radicals (21). Ascorbic acid, the most important antioxidant in plasma, eliminate free radicals in the blood, preventing them from entering into low-density blood lipoprotein (9). However, in one study, Kang et al. (2009) in a study on patients with cardiovascular disease showed that daily doses of 500 milligrams of vitamin C to vitamin E (402 mg per day) and beta-carotene (20 mg per day) had the best performance in reducing oxidative stress (10). Vitamin C directly reacts with O2- and OH °, but its role as an anti-oxidant material has a close relationship with vitamin E. spatial arrangement allows the Vitamin C to have close relationship with vitamin E radicals that produced in the cell membrane. After giving an electron to the vitamin E radical, Ascorbate oxidant into a toxic radical call Dihydroascorbate (SDA) - a less reactive combination – it's a combination with less activity. SDA radical recycled by Dihydroelipoat or through an inadequate reaction comes in Dihydroascorbate (DHA) form. In the presence of GSH, DHA reductase enzyme, catalyzes the Ascorbate reconstruction. In animals, the SDA radical can be directly converted into Ascorbate by using nicotine amide adenine nucleotide dioxide (NADH) as a regenerative effect and SDA reductase enzymes. Vitamin C in particularly was formed to neutralize the free radicals and is effective in watery environments such as plasma. Thus, it can prevent from the red blood cells membrane damage (21). However, most research results suggest that vitamin C can prevent from oxidative stress and of associated complications such as inflammation by anti-oxidation potential increase (8). Also Liu et al (2000), Margaritis et al (2008), are in line (16, 18). Some studies (8, 1). have been reported a high rate in total antioxidant capacity of the blood or muscle response up to 30 minutes to 7 days after the activities. Increased antioxidant capacity, especially in activities such as running a marathon (10, 14), exercise three or pseudo-triple (on the treadmill) (9, 15), have been reported. Due to the increase of this index after long-term activities and sports such as the marathon pseudo-triple could be argued is important variables that is effective in body's antioxidant system stimulating. In addition, the type of activity and body composition factors can influence this indicator response (12).

This findings show that there is a difference between the four groups on maximum oxygen consumption is inactive women. Changes in maximum oxygen consumption between placebo and placebo with training groups, also exercise with supplements been significant. The maximum oxygen consumption in the exercise with supplements group were less than exercise and placebo group. In the meantime, these changes between supplements group and exercise and placebo group as well as exercise and supplements are also significant. Therefore, we can conclude that exercise and placebo groups was better than the other groups. Analysis of the data from the present study showed that four weeks of interval exercise had significant influence (5.24 %) on maximum oxygen consumption. Over the short term place with maximum intensity, high-energy phosphagene metabolism, glycolysis and oxidative metabolism, all of which contribute to ATP regeneration cycle (4). It has been shown that the increase of key regulatory enzymes activity of energy systems, involve in speed enforcement improvement; Therefore seems both stage and speed activity and frequency of training, have effect on performance and compatibility of the enzyme (5). However, multi-stage 30 seconds at maximum power and short rest periods between the stages and the relative contribution of aerobic metabolism increase is probably due to the dynamics of oxygen consumption (6). In this study, after periodic training, VO2max increase amount 5.8 % in training group, may be it is due to the increased oxygen delivery to active muscles or increase active muscle oxygen uptake (increases capillary network and mitochondrial density), as many studies have reported an increase in oxidative enzyme activity, which represents an increase in aerobic capacity (4,7). Also, Bailey et al. (2011) reported a significant increase in VO2peak after two weeks of repeated speed practices (2). No changes in VO2peak in these studies may be due to the use of trained subjects, or short-term training courses (two weeks) and the low number of training sessions. Also, analysis of the data from the present study showed that four week interval training with vitamin C supplementation had a significant effect on maximal oxygen consumption. Researches results (2, 3) of high VO2max in trained individuals compared with the control group are in line with the results of this study. A higher VO2max of training group in the present study show the impact of interval training three days in a week, with 70% of maximum heart rate, nine five minutes sessions, four minutes inactive rest and supplementation index agents and access to favorable terms of physical fitness and cardiovascular risk factors reduction. Previous researches also have shown the physiological superiority of athletes to non-athletes or significant changes in physiological indicators after regular exercise.

The significant effect of exercise on VO2max in patients with severe and moderate aerobic exercise training are observed after 4 weeks. (2). The findings of this study reveal that to achieve the desired level of fitness, training program can be performed during a 4-week was on the agenda of those who wish to get maximum usefulness of their physical activity because of the research is according to the above points.

CONCLUSIONS

The results of the study reveals that relatively intense physical activity, possibly due to excessive production of free bases and anti-oxidative internal resources drain causing increased oxidative stress and oxidative damage against biological macromolecules such as proteins, membrane lipids (Malondialdehyde), the nuclear acids, the results of this study also suggest that three days in a week training, with 70% of maximum heart rate and nine five minutes sessions with four minutes inactive rest, significantly increased serum total anti-oxidation capacity and maximal oxygen is consumed. In addition, the four-week supplementation with vitamin C can promote the prevention of adverse changes in serum total anti-oxidation and oxidative stress damage of the membrane and reduce peak of muscle damage after eccentric exercise.

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