

**Review Article****Effects of an aerobic and anaerobic selective exercise course with green tea consumption on Creatine Kinase Enzyme****Naghmeh Naseri, Shahla Hojjat\* and Asghar Khaledan**

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**ABSTRACT**

**Background and Purpose:** Due to the lack of complete information on the effects of aerobic and anaerobic exercises with the green tea supplementation on Creatine Kinase, this research was conducted to study the effect of an aerobic and anaerobic selective exercise course (resistance) with green tea consumption on Creatine Kinase enzyme in male athletes between 20 to 35 years old.

**Materials and Methods:** 60 male athletes were selected with (age average 26/30 and Standard Deviation 10/73 and weight average 71/68 and Standard Deviation 12/47) and were randomly divided into 6 groups: Group A: an aerobic exercise with Green tea, Group B: an aerobic exercise, group C: an anaerobic exercise (resistance) with green tea, group D: an anaerobic exercise (resistance), group E: only green tea and group F: only performed their own exercises (control). Each 6 groups performed 8 weeks Protocol that includes: Exercises in 2 ways of aerobic exercises, with the intensity 65 to 75 percent of maximal heart rate and anaerobic exercises (resistance exercises), with the weights intensity 60 to 70 percent of a maximum repetition. The experimental group during the research period has received 200mm green tea three days a week. Before the exercises and 8 weeks after the last exercise session, blood samples were taken from subjects in the fasting state.

In all hypotheses, Tukey's range test at the significance level of ( $p \leq 0.05$ ) was used.

**The Results:** According to the results, it can be seen that resting CK changes in six research groups have significant differences. Gyms- Howell's range test results showed that the CK changes in the resistance exercise group with green tea, the green tea group and an aerobic exercise group with green tea have significant differences with two groups of control and resistance exercise. These results indicate that an aerobic exercise with green tea, resistance exercise with green tea and green tea have a reducing effect on the CK values, but their effect is not different.

**Discussion and Conclusion:** Therefore, the results suggest that aerobic, anaerobic (resistance), aerobic and resistance exercises with green tea reduces the CK, in addition, aerobic and anaerobic (resistance) exercises can also increase the CK.

**Key words:** Green tea, anaerobic exercise (resistance), aerobic exercise, oxidative stress, inflammation

**INTRODUCTION**

Oxidative stress with the damage to all kinds of cellular infrastructures will reduce the cellular and physical function, fatigue and muscle damage (Alok et al., 2003), (Anne et al., 2004),

(Atalay et al., 2000). Some researchers believe that by adopting different strategies, in order to inhibit oxidative stress and lipid peroxidation and reduce it, we can prevent from a decline

athletic performance and even we can take steps in order to improve it and increase tolerance toward the exercise (Maria et al., 2003). Oxidative stress occurs when reactive species that are called free radicals, go beyond in a system of the system's ability to neutralize and remove this molecule. Excess free radicals can damage lipids, proteins or DNA of a cell. The creation of these oxygen and nitrogen species will occur regularly as part of normal cell metabolism and will increase under stress conditions (Bloomer, 2007), (Bloomer and Gold farb, 2004). Since the oxidative stress during and after exercise occurs only if the production of reactive oxygen species (ROS) caused by sport go beyond the body's antioxidant defense potential capacity (Konig et al., 2001), (Radak et al., 2001).

Green tea plant is cultivated and grown in Southeast Asia, including China, India, Japan, as well as in many African countries including South Africa (Luximo - Ramma, 2005). Tea leaves have the medical use and the aspect of drinking uses in the society. From 3 thousand years before Christ, traditional Chinese medicine has used the green tea for headaches, body aches, easily digestion, increase defense immunity (safety), to avoid poisoning, as well as an energetic material, and the lifetime (Ferrara et al., 2001). Green tea may prevent the oxidation of cholesterol, LDL and thus reduces the plaque in the arteries, which in turn it improves cholesterol levels and cardiovascular health and epigallocatechin-3-gallat (EGCG) inhibits the activities of Metallo proteinases matrix (MMPs) implicated in the destruction of collagen and thus the effects of repression in cartilage damage in the joints, thus green tea may reduce the severity of rheumatoid arthritis (Babitha et al., 2009). Among the most important group of antioxidants, flavonoids can benoted. Green tea is an important source of flavonoids.

Tea contains a group of flavonoids polyphenolic compounds called catechins and among catechins, the epigallocatechingallate (EGCG) is a powerful antioxidants and the most common and most abundant polyphenols

in green tea (Murase T et al., 2002).Despite scientific advances in today's societies,the green tea consumption as an herbal medicine has made significant progress that has many benefits and used for drinks in the world.

In a research, it is believed that tea is one of the major beverages after water among people and more than two-thirds of the world drink tea (Gupta et al., 2002).

Among drinks rich in flavonoids, green tea is one of the most common (Alessio et al., 2002), (Morillas et al., 2006). Epigallocatechin-3-gallat (is composed of about 95% of the total available catechin. This polyphenols component is the most study cases and the most active in green tea), has the highest power so that its power is respectively 100 and 25 times more than the antioxidant power in vitamins C and E (Gupta et al., 2009). Researchers in a study have confirmed the protective effect of green tea [(600 ml per day) 3 times a day, 200 ml of boiling water with 2 g of dry green tea] in reducing lipid peroxidation in intense resistance exercises (Panza and et al., 2008).Also, they have reported in another study that green tea can increase antioxidant system capacity and reduce oxidative stress caused by strength exercises with an intensity of 60% (1RM) (Jokwo et al., 2011).

Nowadays, they conduct a study entitled the impact of short-term supplementation of green tea on total antioxidant capacity and lipid peroxidation in young women after a severe resistance exercise session. Findings included 14 days of green tea consumption caused an increase in total antioxidant capacity significantly ( $p < 0.001$ ) and a significant reduction of MDA ( $p < 0.01$ ) in the exercise group with the supplementation. However, a severe resistance exercise caused a significant reduction in total antioxidant capacity (TAC) ( $p < 0.04$ ) and a significant increase in MDA ( $p < 0.01$ ) in the exercise group.

Based on the present results, we can conclude that green tea supplementation can reduce unfavorable changes of lipid peroxidation induced by intense resistance exercise through plasma total antioxidant capacity increase

(Ghasemi et al., 2013). In another study entitled the effect of supplementation and anaerobic exercise on markers of oxidative stress that was carried out in water polo athletes, they found that the MDA and the activity of antioxidant enzymes, superoxide dismutase (SOD) and glutathione peroxidase or (GPX) and the catalyze or (CAT) was significantly decreased ( $p < 0.05$ ) and the total antioxidant capacity was significantly increased ( $p < 0.05$ ).

The results of this study based on the findings, it can be stated that taking vitamins C and E may increase the TAC and that is why the destructive oxidizing effects on the lipid membrane of cells and anti-oxidative enzyme activity have been decreased (Didani et al., 2013). While in a research entitled the effects of vitamin supplements consumption (24 mg beta - carotene, 100 mg ascorbate and 800 IU of alpha - tocopherol), were examined. The findings suggest that the use of these vitamins will reduce the fat oxidation (MDA) significantly. The amount of this reduction was higher in people with diabetes that were more susceptible to oxidation of lipoproteins (Anderson et al., 1999).

However, in the research that they carried out, they find the conclusion that aerobic exercises, green tea supplementation consumption and the combination of aerobic exercises with green tea consumption will not create a significant change in the amount of leptin and insulin resistance index. Green tea consumption was significantly decreased body weight and body mass index. An aerobic exercise has significantly reduced body weight, body mass index and percentage of body fat and the combination of the aerobic exercise and green tea caused a significant increase in consuming maximum oxygen and a significant reduction in weight, body mass index and body fat percentage. In conclusion, aerobic exercise and taking supplements of green tea does not effect on the (leptin) and insulin resistance index. Therefore, further study is necessary to achieve more conclusive results (Haghighi et al., 2013). Thus, according to the contradictory results obtained and the paucity of available data on

green tea supplementation during aerobic and anaerobic activities (resistance) and its effect on inflammatory factors and indicators of oxidative stress in athletes after this type of exercises, accurate and controlled studies have not been performed in this context yet, this study was designed.

### Research methodology

The research method is semi-experimental test and the statistical population was male athletes in Tehran, which called for 100 volunteers (participants), respectively. Which among this number ( $N = 100$ ), 60 people were selected purposefully (a questionnaire) by random (simple) as a sample and were divided into 6 groups [5 experimental groups ( $N = 50$ ) and 1 control group ( $N = 10$ )] (Ghasemi et al., 2013) which include:

Group A: Aerobic exercise with green tea

Group B: Anaerobic exercise (resistance) with green tea

Group C: Only drank green tea

Group D: Only aerobic exercise

Group E: Only anaerobic exercise (resistance)

Group F: Only performed their ordinary exercises (control)

A week before the test, measurements of height, weight, age and familiarity with the tests and receiving written consent from were taken from the subjects. In addition, all subjects were in perfect physical and mental health and they had no history of cardiovascular, respiratory and not specific diseases. Before the test, a pre-test was conducted from the subjects and after the end of exercises, meaning after the 8 weeks a post-test was conducted.

200 ml of green tea was given to 30 subjects (2 g of dry leaf green tea in 200 ml of water at a temperature of 80-100°C) as a beverage (panza et al., 2008) and 30 other subjects had no green tea.

After conducting pre-test and post-test, a blood test (fasting) was used. Exercises were 3 days a week for 24 sessions, meaning 8 weeks and 60 minutes duration in each exercise session.

### **Aerobic Exercise**

Aerobic exercises include aerobics, which includes 15 minutes of warm-up exercises (walking, flexibility, stretching, running and doing exercises with bands, lift weights (light), ball, stop performing and doing a series of rhythmic movements that are 35 minutes and 10 minutes cool-down, which generally is 60 minutes of aerobics for three exercise sessions every other day of the week (3 days a week). Doing exercise intensity was between 65 to 75 percent of maximum heart rate (Haghighi et al., 2013), (the new aerobics exercises, Chasten and Jordan, Volume 1), (aerobics - education and its benefits, Veysi and Kashti Dar, Volume 1).

### **Anaerobic exercise (resistance)**

Anaerobic exercise (resistance), which included 10 minutes warm up and then start doing weight exercises, which includes two programs in the form of first to fourth weeks program (the front foot with the device and the back foot with sitting device, chest press with device, emission of behind the shoulder with device, front arm curl with device, back of the arm with device, back stretch with device, crunch with device) that include one set and 12-8 repetition and 2 -1 minute break.

Exercise programs of the fifth to eight weeks (leg press, back foot with the sitting device, the chest with the device, emission of behind the shoulder with device, emission from the side with the device, front arm curl with device, back of the arm standing with a cable, back stretch with device, abdomen crunch with device), which include 1 set and 12-8 reps and 2-1 minute break and doing exercise intensity is from 60 to 70 percent (1RM) (planned reference book of strength exercise: Physiology of power science and work with weights systems / [National Association of strength and fitness in United States], Lee, Volume I).

### **Green tea Supplementation consumption**

In groups that taking supplements of green tea was intended, subjects were asked to maintain their diet for 8 weeks and to brew green tea 3 days per week at a rate of 200 ml (2 g of dry

leaf green tea in 200 ml of water at a temperature of 80-100°C) and it is given to subjects in beverage after exercise.

### **Blood sampling and measurement of research indexes**

Blood sampling was done after 12 to 14 hours of fasting in two stages (before starting the exercises meaning one day before the exercises and after 8 weeks of exercise). In the first stage of blood sampling that was done in the gym of Al-Nabi mosque at 8 am from subjects that blood was taken from the right hand vein of each subject in sitting condition and 5 ml of blood was taken in break condition. In the second stage, the same way was done after the end of eight weeks.

### **How to measure creatine kinase**

For measuring creatine kinase (CK - MB), a serum of Pars test kits, Made in America, micro liter in 340° nm wavelength with the Cubas auto analyzer device and photometric method was used.

To measure Creatine Kinase includes the following steps:

Deutsche Gesellschaft für clinical chemistry (DGKC) Method: (Society of Biochemistry in Germany) and (IFCC), (International Federation of Clinical Chemistry and Laboratory Medicine)

**Test basis:** CK-MB is formed of two subsets of CK-M and CK-B.

At the CK-MB activity that makes up the bulk of the CPK activity and CK-M is a subset of CK-MB that is inhibited by a specific antibody against CK-M and the only CK-B activity that has only half of the CK –MB activity, will be measured.

**Test method:** wavelength: 340 nm, cuvette diameter: 1 cm, temperature: 37 ° C, measurement: photometer with consumption Blank is set at zero. (Stein w et al., 1998), (Moss DW et al., 1999), (Würzburg u et al., 1977).

**Table 3 - 3.** Single solution and the blanks used in the research

Single solution	Blank	Sample
Patient samples	-	40micro liters
Distilled water	40micro liters	-
Mixed solution of 1 and 2	1000micro liters	1000micro liters

After mixing, read the optical absorption after 5 minutes (initial optical absorption) then enables top watch and exactly after 1, 2, 3, 4 and 5 minutes, specify the optical absorption disorders from the minute ago.

### Statistical Methods

To detect and normal data, the Kolmogorov - Smirnov test was used and it was found that the groups are not different from each other. Descriptive statistics were used to calculate the central and dispersion indexes. Inferential statistics were used for one-way ANOVA test and Tukey's range test. Alpha level of significance was considered 0.05. All statistical analyzes were performed by using spss software version 20.

### Research findings

#### Introduction

**Part I:** Study the effect of aerobic exercise and green tea on CK variable

#### The first hypothesis:

**The null hypothesis:** Aerobic exercise with green tea has no significant effect on the CK of male athletes between 20 to 35 years.

The results of ANOVA for comparison of resting plasma CK changes showed a significant difference between changes in four aerobic exercise groups with green tea, aerobic exercise, green tea and the control ( $F_{3,36} = 3.417$ ,  $Sig = 0.027$ ). Table 1 shows the one-way analysis of variance results for the comparison of resting CK changes in four aerobic exercise groups with green tea, aerobic exercise, green tea and control.

**Table 1 -** One-way analysis of variance results from the resting plasma CK changes in four research groups

	Sum of squares	d	Average of Squares	F	Sig
Intergroup	1749.90	3	583.30	3.417	0.027
Within the group	6146.00	36	170.72		
Total	7895.90	39			

According to the results, we can see that resting CK changes in all four aerobic exercise groups

along with green tea, aerobic exercise, green tea and control have a significant difference.

Tukey's test results showed that the aerobic exercise group CK changes with green tea and green tea group are significantly less than the rate of its changes in two groups of control and aerobic exercise.

These results indicate that aerobic exercise has an increasing effect on the amount of CK. However, green tea can decrease the CK amounts significantly either with or without aerobic exercise. In other words, the consumption of green tea could decrease an increasing effect of aerobic activity (aerobic exercise has elevated CK levels) (CK increased in the aerobic exercise group, but no significant increase was observed in exercise and tea groups).

Therefore, we can conclude that the consumption of green tea along with aerobic exercise and the consumption of green tea has a significant effect on the resting levels of CK in men aged 20 to 35 years and the research hypothesis is confirmed.

#### **Part II: Study the effect of anaerobic exercise (resistance) and green tea on the variable CK**

#### The second hypothesis:

**The null hypothesis:** Resistance exercise, along with green tea has no significant effect on the CK amount in male athletes aged 20 to 35 years.

The results of ANOVA for comparison of resting plasma CK changes showed a significant difference between the changes in four resistance exercise groups with green tea, resistance exercise, green tea and the control ( $F_{3,36} = 4.132$ ,  $Sig = 0.013$ ). Table 2 shows the one-way analysis of variance results for the comparison of the resting CK in four resistance exercise groups along with green tea, resistance exercise, green tea and control.

**Table 2** - ANOVA test results from the changes in plasma resting levels of CK in four research groups

	Sum of squares	d	Average of Squares	F	Sig
Intergroup	1558.70	3	519.57	4.132	0.013
Within the group	4526.40	36	125.73		
Total	6085.10	39			

According to the results, we can see that changes in resting CK in each of the four resistance exercise groups along with tea, resistance exercise, green tea and the control have a significant difference. Tukey's range test results showed that the CK changes in the resistance exercise group with green tea and green tea group were significantly less than the rate of its changes in two groups of the resistance exercise and control group, respectively. In addition, there was no significant difference between the CK changes in two groups of Green tea and the combination of green tea and resistance exercise. These results suggest that resistance exercise has an increasing effect on the amount of CK. However, the green tea can significantly decrease the CK values, whether with or without the resistance exercise. In other words, the consumption of green tea can decrease an increasing effect of resistance exercise (resistance exercise has elevated the CK levels). Therefore, we can conclude that the consumption of green tea with resistance exercise and the consumption of green tea on the resting levels of CK in male-aged 20 to 35 have a significant effect and the research hypothesis is confirmed.

### **Part III: The comparison of the aerobic and anaerobic exercise (resistance) and green tea effect on the CK variable**

#### **The third hypothesis:**

**The null hypothesis:** there is no significant difference between the aerobic exercise with green tea and the resistance exercise along with green tea and the green tea on the CK amount in male athletes aged 20 to 35 years.

The results of ANOVA for comparison of resting plasma CK changes showed a significant difference between the changes in

six research groups ( $F_{5, 24.77} = 4.098$ , Sig = 0.008) (there was no homogeneity of variance between four groups, that is the reason why the Welch's amendment has been made). Table 3 shows the one-way analysis of variance results for the comparison of resting CK changes in six research groups.

**Table 3** - ANOVA test results from the changes in plasma resting levels of CK in six research groups

	Statistics	df1	df2	Sig
Welch's amendment	4.098	5	24.77	0.008

According to the results, it can be seen that changes in resting CK in six research groups have a significant difference. Gyms- Howell's range test results showed that CK changes in the resistance exercise group along with green tea, the green tea group and the aerobic exercise with green tea have a significant difference with two groups of control and the resistance exercise. These results indicate that aerobic exercise with green tea, resistance exercise with green tea and the green tea has a decreasing effect on the CK values, but their effect is not different. Therefore, the research hypothesis based on the difference in the effect of aerobic and resistance exercises with green tea will be rejected (Note that in this hypothesis, the focus is on the study of differences in two exercise methods with green tea, not their effect in first and second hypotheses, the effects were studied but in the third hypothesis the effects comparison have been investigated).

## **DISCUSSION AND CONCLUSION**

The research findings are based on that aerobic, anaerobic (resistance), aerobic and resistance exercise with green tea have a reducing effect on the CK. As well as aerobic and anaerobic exercise (resistance) can also increase the CK. CK is an enzyme that involves in the cycle of consumption and tissue energy storage, especially in the muscles. CK is found in very small amounts in blood, but the most amount of it is in skeletal muscle, brain and heart tissue (Burtis et al., 2001). On the other hand, the reduction at the level of muscle activity and

muscle damages can have excitatory effects on the level of CK serum enzyme (Dufou et al., 2001). However, delayed onset muscle soreness in addition to the above symptoms will influence many biochemical variables. Among these, we can refer to an increase in CK and LDH enzymes in the blood and an increase in hemoglobin, hydroxyproline and creatinine in the urine (Talebi Gorgani, 2001). On the other hand, one of the factors, which affect this enzyme, is immobilization stress that has stimulatory effects on the activity level in many biochemical enzymes. Limitation of motion can cause Pathophysiological disorders in the heart muscle and increase the level of CK serum enzyme (Davydov et al., 1999).

As well as reducing oxidative stress, it is recommended to add the use of antioxidants in the diet. Today, much evidence suggests that dietary supplements with plant Bio phenol may be a successful strategy to reduce the risk of pathological conditions related to the overproduction of free radicals and / or prevent their effects (Bonina et al., 2005). A Polyphenolic structure that happens in fruits and vegetables include flavonoids, which cover AntioSanidins, flavonones, and catechins (Yilmaz et al., 2003). Too much emphasis has been done on the beneficial effects of green tea beverages that are a source of catechins, mainly epigallocatechin-3-gallat (EGCG), epigallocatechin(EGC), epicatechingallate (ECG) and epicatechins (EC) ( Erba et al. , 2005). The mechanism of catechin actions includes clearing free radicals, chelation, (Chelating) metal, inhibition of redox - sensitive krypton Trans and Pro-oxidant enzymes, and induction of detoxifying phase II from enzymes (Mcanulty et al., 2004). The consumption of Catechin supplementation can affect oxidative stress through a direct action as well as an influence on other levels of antioxidants in the human body (Zhou et al., 2005).

In a research (Basta et al., 2013) entitled Effect of aloe wood mill on the selected parameters of Pro-oxidant balance cytokine synthesis in boat rower athletes, found that SOD and glutathione

peroxidase activity and the reactive substance concentrations of Thiobarbituric acid (TBARS) in red blood cells was examined. In addition, total antioxidant capacity (TAC) and CK activity was measured in plasma samples, and cytokine concentrations of IL-6, IL-10 in the serum were determined. Before and after Biostimine supplement, the sport is considerably increased the values of SOD, IL-6, IL-10, and TBARS in two groups. However, the levels after exercise and TBARS recovery in recipient Biostimine athletes were considerably lower than the control group. After the consumption of supplemental, TAC was the only variable with a higher significant level in the supplementation group compared to the placebo group. Consequently, we can conclude that Biostimine supplement will decrease post-workout TBARS level with increasing plasma antioxidant activity, but it has no effect on inflammatory indexes. Try another study (Costa et al., 2012) entitled the graph and biochemical changes in the futsal female player through high-intensity intermittent exercise training, they found out that Lough Borough inter mitten shuttle test (LIST) promotes a significant increase in indexes related to lipid peroxidation MDA and lipid hydro peroxides (LOOHs) and CK muscle damage, while it has the tendency to reduce the levels of antioxidants and potential acids and ascorbic.

In a study (Jokwo et al., 2012) entitled the effect of polyphenols single dose in green tea on oxidative Stress markers caused by sport in football players' blood, they found that in both 2 groups, TBARS plasma, UA and TAS are considerably increased after the sport and after a course of 24 hour recovery, it remained high. The SOD activity in red blood cell did not significantly change in response to the muscle and resistance tests, while in both groups, the plasma CK activity increased considerably after 24 hours recovery. Acute consumption of green tea polyphenols (GTP) caused a small but significant increase in total (catechins) available plasma combinations. However, it was specified that GTP did not create a

significant effect on measurable parameters. Therefore, acute consumption of GTP (640 mg) will not reduce the oxidative stress and muscle damage caused by the sport. Findings showed that the physiological pressure on muscle cells incurred because of a session for both exercises probably was the same. It seems that severe activities cause muscle enzymes demonstration particularly the ALD and CK in the blood (Eddington et al., 1994), which may be due to muscle cell damage and an increase in permeability of cell membranes during the activity or after that (Hagh Verdi et al, 2002). They showed that a high level of the skeletal muscle membrane damage would create after sports activity, manifested in the increased activity of CK (Dixon et al., 2003). On the other hand, research shows that apparent and functional and biochemical signs of the pain include limitation of movement, muscle rigidity, muscle strength increase, as well as biochemical signs such as an increase in Creatine kinase and lactate dehydrogenase in plasma (Cheung et al., 2003) and (Clarkson et al., 1982). Based on the above matters, we can say that maybe muscle soreness, loss of muscle activity and muscle damages, immobilization stress (limited mobility), short-term and long-term sports activities and severity of damages, type of supplement, dosage and duration of supplementation, the type of sport and type of exercise are among the factors that have effects on the secretion of the Creatine kinase and stimulate this enzyme.

**CONCLUSION:** aerobic exercise, anaerobic (resistance) and aerobic and resistance with green have a decreasing effect on the CK. As well as aerobic and anaerobic exercise (resistance) can also increase the CK. According to the results, we can say that, aerobic and anaerobic exercise (resistance) with green tea can be useful in reducing the damage caused by CK and prevents an increase in some oxidative and inflammatory stress factors and reduce the damage.

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