The effect of green tea supplementation and climbing to altitude on the serum level of Malondialdehyde and Glutathione Peroxidase In non-professional healthy mountaineer females

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Abstract:

Introduction: Climbing and exercising at high altitudes increase the number of free radicals and reduce the level of antioxidant enzymes in the human body. The purpose of this study was to investigate the effect of three weeks of green tea supplementation before climbing on serum levels of glutathione peroxidase and malondialdehyde in young non-professional female climbers.

Material & Methods: This research was a quasi-experimental study. Twenty-four females (mean age 26.42 ± 7.10 years, weight 58.46 ± 7.96 kg, height 162.50 ± 5.87 cm, and body mass index 22.23 ± 2.67) were recruited voluntarily from a club in Rasht who were randomly divided into two groups: experimental group (EG) and control group (CG). EG consumed green tea daily (20g per day). At the end of the third week, both groups climbed to the Peak of Shah Moallem (3085 meters). In both groups, diet control was performed over three weeks. To measure plasma levels of glutathione peroxidase (GPx) and malonyl dialdehyde (MDA), blood sample were taken in three stages: before green tea supplement consumption, pre and post-climb.

Results: The results showed that compared to CG, the increase of MDA after climbing to the altitude was significantly lower (p= 0/02). Compared to the CG, the amount of plasma GPx increased significantly in the EG after climbing to the height (P=0/001). The results of the research showed the improvement of redox state in mountain climbers after consuming green tea.

Conclusion: The results of this research showed that consuming green tea before climbing to high altitudes can improve the performance of the antioxidant system in climbers.

Keywords: Antioxidant, Free radical, green tea, GPx , High altitude, MDA

1. Introduction

Oxidative stress is defined as a disturbance of the balance between the pro-oxidant and antioxidant components and generating damage. Also, that can decrease the performance of athletes in altitudes Because of the changes it causes in redox (1). Sports such as mountain climbing, biking and skiing are performed at altitude which can stimulate the antioxidant system. But strengthening this system improves sports performance at altitude (2).

The evidence presented strongly suggests that exercises with mild to moderate intensity increase the production of oxidative factors, which the body responds to it with mechanisms such as extensive biogenesis of muscle mitochondria, increased muscle blood supply, and altered fuel consumption patterns[4]. In contrast, endurance exercise of extreme duration and intensity appears to generate much higher levels of free radicals that overwhelm cellular antioxidant defenses and cause tissue damage (3). That Is the result of one of three factors:(1) an Increase in the generation of nitrogen, Through the accumulations of reactive intermediaries: (2) Prejudice of the antioxidant defense system (inhibition of antioxidant enzymes, depletion of non-enzymatic antioxidants): (3) increasing tissue damage due to dysfunction of the antioxidant system (4). It considers that the antioxidant system of the body's deficiency leads to the development of oxidative stress that can damage lipids, proteins, or DNA (5). Altitude exposure can exaggerate the transient increase in markers of oxidative stress observed following acute exercise (6). The generation of reactive oxygen species is typically associated with hyperoxia and ischemia-reperfusion. It was hypothesized that oxidative stress would be increased in subjects exposed to high-altitude hypoxia. (7). High altitude exposure results in decreased oxygen pressure and an increased formation of reactive oxygen and nitrogen species (RONS) which is often associated with increases in oxidative damage to DNA. Exposure to high altitude decreases the activity and effectiveness of the antioxidant enzyme system (8). The antioxidant system of the organism can be divided into enzymatic and non-enzymatic. The major components of the enzymatic antioxidant system are superoxide dismutase, Catalase, and glutathione peroxidase, which avoid the accumulation of radicals O^{2-} and H_2O_2. The non-enzymatic antioxidants include compounds produced “in vitro” such as reduced glutathione (GSH), ubiquinone, uric acid, and transition metal transport proteins (transferrin) (8, 9). The body makes enough antioxidants in normal situations, but it isn't enough in intensive exercises or at high altitudes (2).Green tea is obtained from the green leaves of the camellia plant, which contains large amounts of polyphenols (10). Consumption of antioxidant substances reduces the damage caused by oxidative agents. This effect improves exercise performance and health in athletes. Eating foods rich in antioxidants can be a natural solution to reduce the negative effects of oxidants (11) For example aerobic training with or without green tea supplement can decrease triglyceride, low-density lipoprotein, blood pressure, and heart rate (HR) significantly ($P < 0.05$)(12) . Green tea is one of the strongest antioxidant drinks in the world (13, 14).

In a research, it was shown that tocopherol and carotenoids had no effect on the activity of glutathione peroxidase and catalase, but Green tea extract significantly reduced plasma iron versus baseline(15). The exact mechanism that causes the antioxidant agent to prevent damage caused by oxidative agents is not clearly known and requires further research (15, 16). The purpose of this research is to determine the effect of green tea supplementation and climbing to altitude on the serum level of GPx and MDA in non-professional healthy mountaineer females.

2. Materials & Methods

Subject

In this quasi-experimental study, Twenty-four females (age 26.42 ± 7.10 years, weight 58.46 ± 7.96 kg, height 162.50 ± 5.87 cm, and body mass index 22.23 ± 2.67) were recruited voluntarily from a mountaineering club in Rasht. The criteria for entering people were to be healthy and not to use some drugs, cigarettes, and alcohol, who were randomly divided into two groups: the experimental group (EG) and the control group (CG). The experimental group consumed one cup of green tea daily (20g per day) (17). All members signed the consent form.

Exercise training and supplementation

In a period of three weeks, participants drank green tea (125 cc of solution containing 20 grams of green tea) that has antioxidant effects. The food plan of both experimental and control groups was controlled based on the recommended percentage of healthy food plan, which included %55-%65 carbohydrates, %15-%30 fat, and %20-%25 protein. Blood samples were taken three times. The first one was done at 7 am (before the start of study). 12 hours later and after climbing Shah Moalem peak in the evening of the same day, the next blood samples were taken. Climbing to the top of the mountain was done in October 2023 with a temperature of 15 degrees Celsius at the base of the mountain and 2 degrees Celsius at the top and with a humidity level of 30% to 50%. This climb lasted about 8 hours. Blood was taken three times by nurse, and stored in double-walled test tubes in an ice cube tray and delivered to the laboratory of Shahid Beheshti University.

Measurements

GPx activity is carried out by colorimetric method with glutathione peroxidase activity assay kit (Zell Bio Co. Germany). Malondialdehyde (MDA) was measured by calorimetric method and using TBARS Assay Kit (Zell Bio Co., Germany). A list of high antioxidant foods was provided to the participants to avoid during the study (eg garlic, olive, red & and orange fruits, sour lemon, and soya) (18).

Statistical Analysis

To describe the values of the variables, the mean and standard deviation were expressed, and for statistical analysis, t -test, Man-Whitney U and covariance tests were used. Statistical analysis was performed using SPSS software at a significance level of (0.05).

3. Results

The dependent variables of the research were MDA (a marker of oxidant in the body) and GPx (one of the antioxidant enzymes) in the blood serum of healthy non-professional mountaineer females. For statistic explanation was used t- test for GPx , and is used Mann–Whitney U test for MDA. Descriptive information about the EG and CG groups showed that in Table 1.

Table 1: Descriptive information f control group (CG) and experimental group (EG)

Variable	Pre-Test		Post-test	
	CG	EG	CG	EG
Age (Year)	24.42 ±6.12	28.42 ±7.70	----	----
Height (cm)	1.62 ±0.06	1.63 ±0.04	----	----
Waist (cm)	79.16 ± 7.54	79.16 ± 7.54	----	----
Waist-to-hip ratio	0.75 ± 0.04	0.77 ± 0.04	----	----
Weight (kg)	59.53 ±7.97	59.53 ±7.97	56.91 ±8.14	58.41 ± 7.68
body mass index	22.14 ± 2.86	22.32 ± 2.59	21.75 ± 2.86	21.89 ± 2.49

Changes of MDA and GTX was shown in Figure 1 & 2.

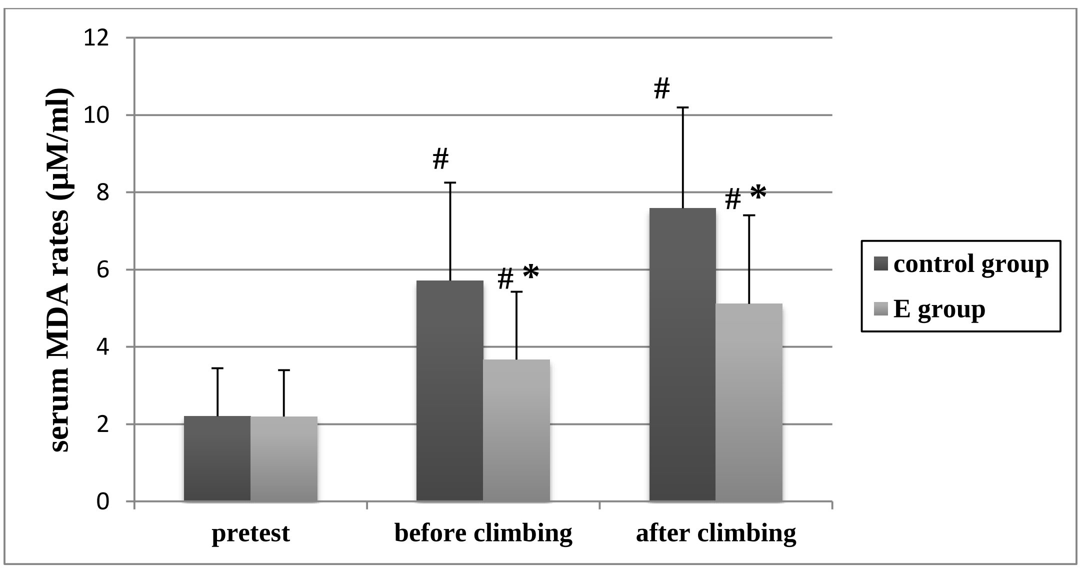

Figure 1. MDA changes : The symbol * indicates the difference between the two control and experimental groups in each of the stages of blood sampling, and the symbol # indicates a significant difference with the baseline values before consuming green tea.

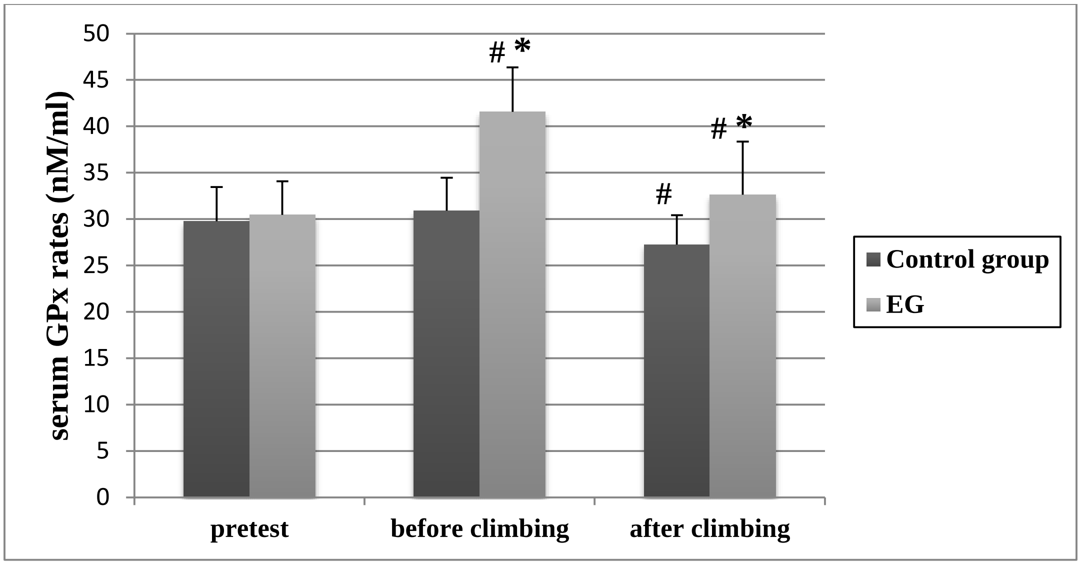

Figure 2. GPx changes: : The symbol * indicates the difference between the two control and experimental groups in each of the stages of blood sampling, and the symbol # indicates a significant difference with the baseline values before consuming green tea.

According to the results, three weeks of green tea supplementation has reduced MDA compared to the control group (P=0.02), and significant increase in plasma level of GPx in two measurements on the day of climbing in the experimental group (p=0.013).

4. Discussion

Many physiological changes occur in the body according to the intensity and type of exercise and conditions and the environment condition in which they exercise It has advantages and disadvantages (19) .Some of the attractive and popular Sports venues are mountains and heights in many sports fields, including mountaineering (20). Two factors are important in high-altitude sports training, the reduction of oxygen pressure and the increase of oxidizing agents (21). According to our findings, climbing up to 3000 meters altitude after consuming green tea for three weeks can decrease the plasma level of MDA and increase GPx. Aghajani et al. showed that aerobic and resistance training with or without supplementation (saffron) consumption can be regarded as an effective method to improve the peroxidase and antioxidant balance (22). The basic issue of oxidation pressure and redox changes that occur at altitude causes changes, which in this research was related to changes in factors such as MDA. MDA is an active and highly reactive aldehyde compound. It is produced in the human body from the peroxidation of unsaturated fatty acids (23). Amount of peroxidation of fats and use it as an indicator to measure the level of oxidative stress by measuring the amount of MDA in different biological samples. MDA itself affects the function of molecules and ultimately the function of the cell (24).

It considers that the antioxidant system of the body's deficiency leads to the development of oxidative stress that can damage lipids, proteins, or DNA (4). Altitude exposure can exaggerate the transient increase in markers of oxidative stress observed following acute exercise (5). The generation of reactive oxygen species is typically associated with hyperoxia and ischemia-reperfusion. It was hypothesized that oxidative stress would be increased in subjects exposed to high-altitude hypoxia (6). High altitude exposure results in decreased oxygen pressure and an increased formation of reactive oxygen and nitrogen species (RONS) which is often associated with increases in oxidative damage to DNA. It decreases the activity and effectiveness of the antioxidant enzyme system (7).The generation of ROS is derived from the contradictory roles that oxygen plays in metabolism. Oxygen is essential. On the other hand process of oxygen reduction to water can generate ROS as intermediates with the potential to cause damage (25).Oxidative stress occurs as a consequence of aberrant reduction-oxidation (redox) control when there is the overproduction of ROS and/or insufficiency of the antioxidant defense mechanisms (25). To protect biological systems from free radical toxicity, several cellular antioxidant defense mechanisms keep the production of ROS in check, including enzymatic and non-enzymatic pathways (26).

The GPx have grown up into a huge family of enzymes that prevent free radical formation from hydroperoxides, and thus are antioxidant enzymes, but they are also involved in regulatory processes or synthetic functions (27). The reaction mechanism has been investigated by the density functional theory and nuclear magnetic resonance of model compounds mimicking the reaction cycle. The resulting concept sees a selenate oxidized to a selenic acid (28).Green tea, derived from Camellia sinensis, is one of the most widely consumed beverages in the world especially in Asia. The main constituents of tea leaves are polyphenols (29).The antioxidant activity of polyphenolic compounds is associated with the capability of inactivation of reactive radical species. Neutralization occurs when an antioxidant transfers its electron and/or hydrogen atom to the radical. The scavenging of free radicals by phenolic compounds can follow four chemical pathways (30).

It has been found that the level of MDA increases at altitude, and on the other hand, the antioxidant enzymes (glutathione peroxidase) decrease. GPx is the general name of a family of enzymes with peroxidase activity, whose main biological role is to protect organisms against oxidative damage (31). The biochemical function of the glutathione peroxidase enzyme is the reduction of lipid hydroperoxides to the corresponding alcohols and the reduction of free water hydrogen peroxide (10). It is produced naturally to protect the body from free radicals, and in this research (32). It was determined that its serum level decreased at altitude. Green tea is impressive in the level of MDA in blood serum (33). Flavonoids of green tea affect health (34). Polyphenols of green tea are strong antioxidants. Their mechanism is adjusting the activities of cells in the signaling way of Protein kinase and Lipid Kinase (35). Climbing to altitude increases the level of markers of oxidative stress (31). Climbing can be a reason for increasing oxidative stress but it can be adjusted by nutrition. consuming antioxidant food or some vitamins and antioxidant supplements (36).

Consuming green tea can be useful for encountering increasing oxidative stress in climbing to high altitudes as one of the strong antioxidants (37). green tea extract exhibits extremely potent protective effects against senescence-mediated redox imbalance in the livers and kidneys of mice by inhibiting oxidative damage of lipids and proteins and increasing the activities of antioxidant enzymes in the body (38).

However, in inconsistent studies, it has been shown no effects were noted in serum levels of carotenoids and tocopherols and GPx and catalase activities. Green tea extract significantly reduced plasma iron versus baseline (16).

The exact mechanism of an antioxidant supplement to reduce the damage by free radicals is unknown and it needs more study (15). One of the limitations of this study is the small sample size, focusing only on the gender of females in a certain age group.

5. Conclusion

In conclusion, the result of the investigation according to the mentioned statistical results and the significance level was found that the laboratory and statistical results show that high altitudes and climbing them can be the reason for increasing the oxidative stress markers like MDA in healthy non-professional mountaineers' females and it's proved that can be adjusted by controlling and enhancing antioxidant enzymes like as GPx after consuming green tea during the 20 days before climbing. In this study, the consumption of green tea is recommended for being useful and effective for regulating the oxidative function of the body and thus establishing homeostasis in the conditions of sports training at high altitudes.

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