

The effect of chamomile extract on antioxidant indices in young female futsal players

Malihe Jafari*

MSc student of Exercise Physiology, Department of Exercise Physiology, Faculty of Sports Sciences, Allameh Tabataba'i University, Tehran, Iran

Bakhtyar Tartibian

Professor of Exercise Physiology, Department of Exercise Physiology, Faculty of Sports Sciences, Allameh Tabataba'i University, Tehran, Iran

Seyed Morteza Tayebi

Associate Professor of Exercise Physiology, Department of Exercise Physiology, Faculty of Sport Sciences, Allameh Tabataba'i University, Tehran, Iran.

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Abstract

Background: Limited research has investigated the use of chamomile as an antioxidant supplement in team sports. **Purpose:** The aim of this study was to investigate the effect of chamomile extract on antioxidant indices in young female futsal players. **Methods:** 20 young female futsal players (Average age: 22.08 ± 5.03 years, weight: 54.4 ± 8.62 kg, and body mass index: 20.56 ± 2.6 kg/m²) were divided randomly into two groups, experimental group (10 people) and control group (10 people). The experimental group consumed 1500 mg/day of the supplement for one week, while the control group received a placebo. Both groups were nutritionally controlled during this period. After the end of the intervention period, two simultaneous futsal matches were held for two groups (two teams of 5 players in each match). Anthropometric characteristics were evaluated in resting conditions and biochemical markers were measured before the intervention, immediately after the competition, and 30 minutes after the competition. Data analysis was performed using the repeated measurement variance test. The level of significance was considered at $P < 0.05$. **Results:** The results showed a significant increase in SOD levels ($P < 0.05$) and a significant decrease in CAT levels ($P < 0.05$) immediately after the competition in the supplement group, but this significance was not seen 30 minutes after the competition. **Conclusion:** One-week consumption of chamomile improves antioxidant defense in young female futsal players. Conducting studies with long-term follow-up to confirm these findings should be considered.

Keywords: Chamomile, antioxidant, futsal match, catalase, superoxide dismutase

* **Author's e-mail:** malihe.jaafari@gmail.com (**Corresponding Author**);
ba.tartibian@gmail.com, tayebism@gmail.com

INTRODUCTION

Futsal is one of the most popular sports in Iran, and Iran has been ranked among the top 10 teams in the world (Farhani et al.2019). Female futsal is among the fastest-growing sports worldwide, having more than 12 million players spread over 100 countries. However, there is less information about the physical demands on female futsal players than on male futsal players in the literature (Castillo et al.2022). Although futsal has similarities with other football disciplines, the unlimited number of substitutions allows a higher intensity during match play because players have the opportunity to recover during substitutions and time-outs. Additionally, the low number of players (i.e., 4 field players plus a goalkeeper) and the reduced available space for play (i.e., 40 × 20 m) demands that elite futsal players must possess a combination of excellent physical, technical, and tactical skills. From a physical point of view, futsal performance has been associated with high levels of agility, muscle power, repeated sprint ability, and maximal oxygen uptake (60 ml/kg-1 min-1) (Á Lopez-Samanes et al.2021). The sprint, jump, and agility tests were sport-specific functional tests, validating the match performances of basketball and soccer athletes. They could assess the power, speed, and agility of the lower extremities for a specific sport (W.-D. Chang et al.2020). Several studies have found that the incidence of injuries in futsal is 2 to 6 times that of football, which is considered a high-injury sport (A Pourrahim et al.2021). Immediate and short-term changes are induced by a single futsal match on oxidative, inflammatory, muscle damage, and antioxidant markers (K. Kaliarntas et al.2023). The increase in oxygen consumption during exercise promotes a large leakage of FR in the mitochondria and then results in an antioxidant reaction (W Souissi et al.2020). When the production of free radicals overwhelms the body's antioxidant defense system, oxidative stress occurs (T. M. Delarosa et al.2023). Oxidative stress reduces strength and performance; mechanically, reactive oxygen species (ROS) can speed up skeletal muscle fatigue by reducing calcium sensitivity (H Arazi et al.2021). However, the antioxidant system of these cells can effectively protect them against severe damage. Two major classes of endogenous protective mechanisms work together to ameliorate the harmful effects of oxidants in the cell. The primary enzymatic antioxidant system found in cells includes superoxide dismutase (SOD), glutathione peroxidase (GPX)

and catalase (CAT) (M Dekany et al.2008). In this regard, Fatoros et al. (2010) investigated the levels of oxidative stress and antioxidant status of 20 football players during the return to the initial state; presented the first data on oxidative stress conditions after a football game. The findings showed that oxidative stress is strongly aggravated by the game and as part of the inflammatory response caused by exercise, it is associated with a significant worsening of anaerobic performance for 72 hours. These cases have led to the use of alternative products for classic antioxidants that are able to modulate redox responses without reducing some beneficial sports adaptations. Hundreds of natural polyphenols are present in edible plants. Most polyphenols have free radical-scavenging capacity. Some polyphenols may enhance sports performance and facilitate the adaptation to regular exercise by reducing exercise-induced muscle damage (M Gelabert-Rebato et al.2019). Due to the complications and adverse effects of chemical drugs, the tendency to use medicinal plants has increased significantly in the world today. Among these plants, we can mention chamomile with the scientific name *Matricaria chamomilla* L (N Mirazi et al.2019). Chamomile (*Matricaria chamomilla* L.) is a member of the Asteraceae family and contains various phenolic compounds (a polyphenol group member) and flavonoids (BARCIN-GÜZELDERE et al.2022). Chamomile is best known for its anti-inflammatory, antioxidant, anti-cancer, neuroprotective, anti-allergic, and anti-microbial properties (Saara Ahmad et al.2022). This is because of its content of biologically active compounds including essential oils and several polyphenols (NAN Hanafy et al.2022). Polyphenols with antioxidant properties are crucial among these nutrients for boosting endurance capacity by enhancing mitochondrial biogenesis and fatty acid intake and reducing oxidative stress. By eliminating free radicals, such as superoxide anions, polyphenols also aid in maintaining the body's immune system (Katsuhiko Suzuki 2021). There are five main groups of phenolic compounds in chamomile extract: coumarins, phenylpropanoids, flavones, flavonols, and flavanone (Z Asadi et al.2020). According to previous studies, no study has measured the effect of chamomile plant consumption on antioxidant levels after a futsal game, especially in women, and most of the therapeutic effects of chamomile have been investigated. In this way, improving the existing knowledge in relation to nutritional knowledge and the response of the mentioned indicators after consuming the chamomile plant as a strong and

natural source of polyphenols after a futsal game which was the aim of this research seems to be an important issue that can be used to promote health and performance, especially in female futsal players.

METHOD

A randomized, double-blind, placebo-controlled experiment was used. 20 young female professional futsal players from Tehran province clubs (age: 22.08 ± 5.03 years, weight: 54.4 ± 8.62 kg, and body mass index: 20.56 ± 2.6 kg/m²) volunteered to participate in this study (Table 1). The sample dimension analysis was performed using G*Power software. Based on prior analysis, we adopted a power of 0.7, $\alpha = 0.05$, and an effect size of 0.8. From these values, an N of 20 subjects was calculated. The criteria for entering the subjects into the research were: female gender with an age range of 18 to 29 years, professional futsal player with at least 3 years of experience in club, provincial or national, and national competitions, not using anti-inflammatory drugs, no allergy to chamomile, no nutritional diet, no smoking. The exclusion criteria for this study were: suffering from any respiratory diseases or joint and bone injuries, taking some antidepressants such as fluoxetine, as well as alcohol, aspirin, warfarin and heparin (due to drug interactions with chamomile capsules), fail to complete the period of receiving supplements and having any underlying disease and intolerance to chamomile, taking stimulants, taking painkillers and anti-inflammatory drugs, using a diet outside of the researcher's recommendation. All participants provided signed informed consent after being informed of the testing and training procedures to be performed during the study. The study was approved by the Allameh Tabatabai University Ethics Committee (number IR.ATU.REC. 1402.013).

Table 1: Participants' characteristics

Group	Body Fat (%)	BMI (kg/m ²)	Weight (kg)	Height (cm)	Age (year)
Supplement	18.86±6.34	20.54±2.8	55.29±10	163.8±5.71	20.6 ±3.68
Placebo	19.9±4.9	20.57±2.45	53.57±7.39	161.3±3.8	25±5.39

Procedures

At the beginning and in basic conditions, the study variables such as weight, height, body mass index (BMI), and body fat percentage (FFM) were measured using the body composition 365 x_contact device made in South Korea, and in order to determine the biochemical indicators of the subject Comment (SOD and CAT) blood sampling was done from all the subjects. It is worth noting that, after completing the consent form and questionnaires about food and medical-sports knowledge and explaining the research process, the diet of the subjects was standardized from 72 hours before the start of the study until its end. The subjects were advised to refrain from taking any drugs, food supplements, or tobacco. Also, the day before the presence of the subjects to measure the variables, they were advised to fast and not consume caffeine and heavy activity in order to reduce the error of measuring the percentage of muscle and fat by the body analyzer. Then the subjects were randomly divided into two supplement groups (10 people) and a placebo group (10 people). For 7 days, the supplement group used chamomile capsules containing 500 mg of dry chamomile extract three times a day, and the control group used placebo capsules of the same color containing 500 mg of breadcrumbs made by Karaj Medicinal Plants Research Institute. To minimize the effect of the intervention variable, supplementation was done in a double-blind manner. After one week, the subjects appeared in the laboratory as on the first day. After measuring the anthropometric variables, a standard breakfast that included 1 tablespoon of peanut butter, half a tablespoon of honey, 60 grams of bread, and 2 dates along with tea was consumed. Then, the subjects competed in two futsal matches with two 20-minute halves in four teams of five, and were only allowed to drink water during the competition. In order to measure the biochemical variables, immediately after the end of the race and 30 minutes after the race, blood was taken from all subjects on two occasions.

Official match protocol

The official match was held with a special warm-up stage for the teams for 20 minutes and then the game was played in two 20-minute halves with useful time conditions and a 15-minute break between the two halves with the presence of referees and spectators.

Blood Sample Assessments

In this study, 5 cc of blood was taken from the subject's arm vein in three stages. Before blood sampling, the subjects were advised not to do any heavy physical activity and consume antioxidant substances as much as possible for 72 hours before the tested protocol as well as blood sampling and basic measurements. Blood samples were collected before taking the supplement, immediately after the race, and thirty minutes after the race. Then the blood samples were placed in tubes containing separating gel and centrifuged at 3000-3500 rpm for 10 minutes at 4°C. The levels of superoxide dismutase (SOD) and catalase (CAT) enzymes were measured by ELISA method using Zelbio Germany research kit for human samples with sensitivity of 1 and 0.5 U/ML respectively.

Statistical Analysis:

Data are presented as mean \pm SD. The normality of each variable was tested using the Shapiro-Wilk test. Then, the changes of each of the indicators during different stages of measurement using the repeated analysis of variance test (to compare data within groups) and Bonferroni's post hoc test at the significance level of $\alpha=0.05$ using SPSS version 26 statistical software.

RESULTS

The study found that, in the comparative analysis, for both CAT and SOD variables, the results of the Mauchly's test indicate the establishment of the hypothesis of sphericity ($p=0.95$) and ($p= 0.072$). Therefore, to check the difference between the measured times, Sphericity Assumed corrective tests were used. The results of this test showed that there is a significant difference between the two times of blood plasma measurements before supplementation and immediately after the competition, ($p=0.04$) and ($p=0.02$) respectively. These results are also shown in Table 2, Fig1 and Fig2.

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Table 2: Results of analysis of variance with repeated measures

	df	Mean Square	F	Sig.
CAT*Group	2	18.79	3.48	0.04*
SOD*Group	2	40.96	4	0.026*

* Significant at $P \leq 0.05$ level

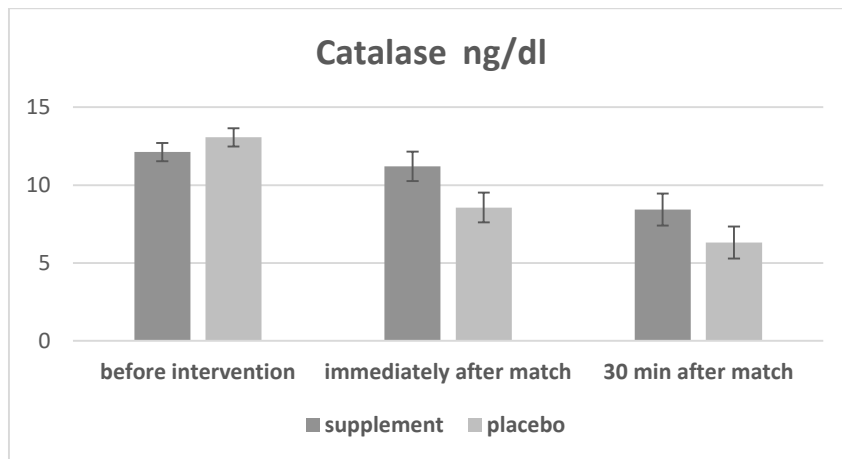


Fig 1. Changes of CAT levels of two supplement and placebo groups before, immediately, and 30 minutes after the competition; there is a significant difference immediately after match.

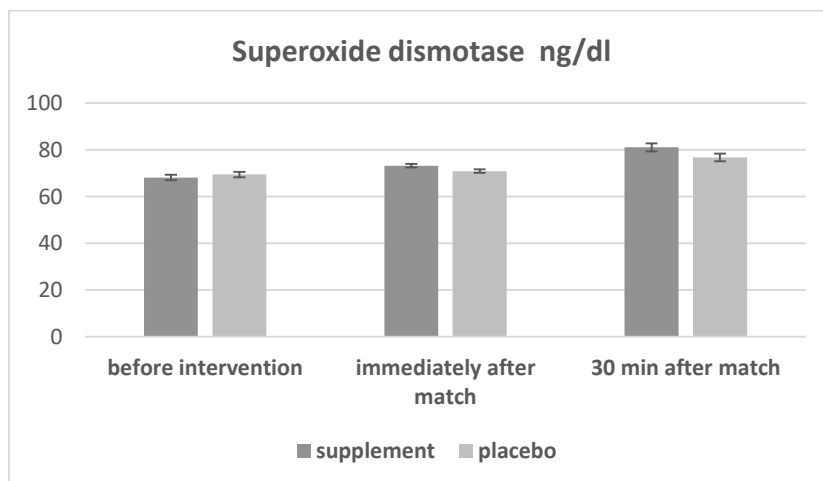


Fig 2. Changes of SOD levels of two supplement and placebo groups before, immediately, and 30 minutes after the competition; there is a significant difference immediately after match.

DISCUSSION

This study was designed with the aim of investigating the effect of chamomile supplement consumption on the antioxidant indices of young female futsal players. The main findings of this study showed a decrease in the amount of catalase enzyme immediately after the race and 30 minutes after the race in both supplement and placebo groups. The difference between the two groups in the first measurement, i.e. immediately after the race, was significant. In a study on female futsal players, it was shown that CAT increased immediately after the match and returned to the initial state 24 hours after the match. (Souglis et al.2023). In this study, all participants had to be in the follicular phase of their menstrual cycle (2 to 8 days after the start of menstruation), that is when the concentration of estrogen and progesterone is low. The reason for the lack of similarities between this study and the present study may be due to the difference in the follicular phase in the present study. A study by Casey et al.2012 on football players and post-match reduction of catalase showed that acute exercise leads to the production of ROS and thus worsens lipid peroxidation, which increases MDA levels, possibly through exhausting the antioxidant defense system. It is reflected that it is consistent with the results of the present study. Hodgson et al.1975 in a study showed that following intense and high-pressure aerobic exercise, the activity of antioxidant enzymes decreases during the increase of activated oxygen species and oxidative stress may be related to their greater use. against free radicals and on the other hand due to the restriction of antioxidant enzymes by activated oxygen species. In a study, Tartibian et al.2013 showed that catalase gene expression increased significantly after exercise in female athletes, which was not consistent with the results of the present study. Considering that the amount of production of free radicals depends on the intensity, duration of activity, and adaptation to sports activity, probably the difference in intensity and type of sports activity is the reason for this disparity. The studies of other researchers in this field show that the longer the activity is done and the intensity is increased, the level of free radicals will increase

subsequently. In a study on soccer players, Akbarpour et al. (2014) showed that intense sports activity causes an insignificant increase in the level of lymphocyte catalase in young soccer players, which may be due to the adaptation of the lymphocyte catalase enzyme of soccer players to intense activity. Hajizade et al. (2013) tested the effect of 16 weeks of intense cycling training on 75 people in the off-season on oxidative stress and antioxidant capacity of male road cyclists. The results showed that an increase in ROS and a decrease in antioxidant capacity occur after this type of exercise. Research studies show that sports training changes the antioxidant activity of tissues according to the type of sports protocol, the volume of training, and the presence of rest periods between training programs. For example, intense training with insufficient rest leads to the stimulation of neutrophils. Neutrophils can cause the production of ROS, which ultimately leads to the creation or increase of oxidative stress and, as a result, reduces the antioxidant defense (Rami M et al.2018). It is possible that the reason for these contradictions is related to the effects of aerobic exercise in increasing resistance to oxidative stress and reducing the need for CAT enzyme for antioxidant defense and activation of other antioxidant pathways. MT Goodarzi et al. (2019) in their study showed that, in oxidative conditions, if the body's homeostasis system is not able to compensate, the oxidative stress causes a decrease in the antioxidant capacity and a decrease in the activity of catalase enzymes. According to the fact that in the present study, a lower catalase drop was observed in the supplement group, which probably means that the use of chamomile as a polyphenolic supplement was effective in helping the antioxidant system. The present study showed an increase in the amount of superoxide dismutase enzyme immediately after the race and 30 minutes after the race in both supplement and placebo groups, and the difference between the two groups was significant in the first measurement, i.e. immediately after the race. In a study on inactive girls, Faizi et al. (2022) showed that immediately after performing two intense and moderate resistance exercise protocols, there was no significant change in the activity of the SOD enzyme, but after two weeks of taking an antioxidant supplement, there was a significant increase in the activity of this enzyme. GH Jahani et al. (2010) in a study that evaluated the effect of the implementation of bulk and rest protocols on the levels of superoxide dismutase in young men, reported a significant increase in superoxide dismutase levels. Yeylaghi Ashrafi et

al.(2016) in a review study, showed that the implementation of an intense exercise activity leads to a non-significant increase in the expression of the superoxide dismutase gene or no significant change in it and they stated that this kind of inconsistency can be considered due to the difference in the researchers' reports of the two terms compatibility and the subsequent response of compatibility so that some researchers have used stimulatory protocols to check the response of stress indicators to a bout of exercise following adaptation to exercise, while other researchers have reported resting levels of this index following adaptation to exercises. G Fisher et al. (2011) following a 20-minute HIIT session, consisting of four minutes of pedaling at an intensity of 15% of maximum anaerobic power and 30 seconds of activity at an intensity of 90% of maximum anaerobic power for four repetitions; have observed an increase in lymphocyte SOD. In Parker et al.'s study (2016), a significant decrease in superoxide dismutase was observed following a HIIE session. The inconsistency in the findings of the current research and the results of the study by Parker and others can also be caused by the difference in the activity protocol, age, or physical fitness level of the subjects. Sadighara et al. (2013) In a study, showed that the chamomile plant has both antioxidant and anti-inflammatory effects. In a study that examined the antioxidant properties of chamomile essential oil and pure azulene essential oil, Asgari et al. (2001) showed that chamomile essential oil has the highest antioxidant effect and inhibits lipid peroxidation. In a study conducted by Asgari et al. (2001), it was observed that the activity of antioxidant indices in the liver cells of rats increased significantly after consuming chamomile extract. In Alouei et al.'s (2017) study, daily consumption of chamomile leaves aqueous extract solution (200 mg/kg body weight) for 8 weeks significantly increased the activity of superoxide dismutase enzyme in type 1 diabetic male Wistar rats compared to the diabetic control group. which indicates the positive effect of chamomile leaf extract on reducing oxidative stress. Therefore, in the present research, the extract of chamomile probably prevented the start of the destructive process in the muscle tissue through its antioxidant effect. The increase of plasma SOD indicates that the oxidation and antioxidant balance partially moves towards anti-oxidation. Maintaining a balance of oxidation and anti-oxidation in a state that favors anti-oxidation is beneficial for the overall health of the body. Therefore, the increase in

plasma SOD is considered as a positive result in this study, which showed a significant increase in the chamomile supplement group compared to the placebo group after the competition. Since the effect of chamomile extract consumption on female futsal players has not been investigated so far, supplement dosage, intervention duration, and individual characteristics are probably some of the significant intervening factors that are suggested to be considered in future studies.

CONCLUSION:

The current findings indicate that the consumption of chamomile extract can increase antioxidant defense after a futsal match and reduce oxidative conditions. The use of chamomile as a food supplement in futsal sports may reduce the adverse effects of oxidative stress created during the competition and maintain the performance of players at an optimal level. However, in order to clarify the exact mechanisms of these changes, we need more detailed and controlled studies in the future.

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