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The effects of natural SOD administration on muscular and hepatic enzyme profile in high-performance athletes: a retrospective clinical study

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Abstract

Background. Antioxidant supplements, such as natural Superoxide Dismutase (SOD), have been studied for their essential role in reducing oxidative stress, preventing muscle damage, and enhancing post-exercise recovery by protecting muscle and liver cells from the damage caused by reactive oxygen species (ROS) generated during intense physical exertion.

Aims. The aim of the study is to evaluate the impact of natural SOD administration on the reduction of oxidative stress and on the modulation of the muscular and hepatic enzymatic profile in elite athletes.

Methods. The retrospective clinical study was conducted over two distinct periods, assessing the effects of natural SOD administration on elite soccer players. The study subjects were divided into an experimental group (EG) and a control group (CG), with a total of 42 participants. The CG included 16 subjects, while the EG consisted of 26 subjects. Each group was evaluated before and after the treatment, which lasted for a period of 2.5 to 3 months. Biochemical evaluations included tests for parameters such as AST (Aspartate Aminotransferase), ALT (Alanine Aminotransferase) and creatine kinase (CK). Statistical analysis was performed using KyPlot 6.0 software, applying non-parametric tests for non-normally distributed and paired data, with a confidence level of 95%.

Results. The comparative analysis between the control and experimental groups revealed significant differences in the enzyme levels of AST (TGO), ALT (TGP) and creatine kinase (CK), with moderate effect sizes. This fact suggests a potential positive impact of the antioxidant treatment, more pronounced in female athletes. In contrast, no statistically significant changes were observed in the male subgroup. The intra-group analysis indicated significant decreases in AST, ALT and CK levels within the experimental group, reflecting a protective effect on liver function and a reduction in muscle stress following the treatment, thereby confirming the efficacy of the antioxidant intervention ($p < 0.05$). Although no values exceeding the normal reference ranges were explicitly mentioned, it is important to note that, in general, standard concentrations for AST, ALT and CK may vary depending on the specific methodologies employed by each laboratory. Elevated levels of AST and ALT may indicate liver damage, while increased CK values can result from intense physical exertion or muscle injury.

Conclusions. The results of the comparative analysis between the control and experimental groups confirm the hypothesis that antioxidant treatment has a significant impact on the activity of AST (TGO), ALT (TGP) and creatine kinase (CK) enzymes. This treatment has a protective effect on liver function and reduces muscle stress, with a more pronounced impact observed in female athletes.

Keywords: antioxidant supplementation, CK, biochemical parameters, elite soccer players.

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Introduction

The effects of antioxidant supplementation on elite athletes have been extensively studied in recent years, given the essential role these compounds play in mitigating oxidative stress, reducing muscle damage and enhancing post-exercise recovery processes. Reactive oxygen species (ROS), generated during high-intensity physical activities, can cause significant damage to muscle and liver cells, thereby increasing oxidative stress and triggering inflammatory responses. In this context, antioxidant supplements such as natural Superoxide Dismutase (SOD) are being investigated for their potential to boost athletic performance by protecting cells against the harmful effects of ROS and supporting recovery mechanisms (Clemente-Suárez et al., 2023).

Antioxidant supplements play an important role in decreasing oxidative stress, inflammation and muscle damage. They contribute to improved recovery and athletic performance in soccer players. However, further research is necessary to clarify the underlying mechanisms and the long-term benefits of such interventions (Poulios et al., 2024). Studies have shown that antioxidant administration can have a considerable influence on the enzymatic profile of athletes by lowering muscle damage markers, such as creatine kinase (CK), and improving the capacity of the body to cope with oxidative imbalance (Arent et al., 2010). In a study conducted by Gonçalves et al. (2022), it was demonstrated that supplementation with phenolic compounds, including natural SOD, contributes to the diminution of oxidative damage and to the enhancement of post-exercise recovery, positively impacting physical performance. Furthermore, Hammouda et al. (2011, 2012) observed an increase in total antioxidant levels in the blood following short-duration, high-intensity exercise, highlighting the role of antioxidants in reducing oxidative imbalance and preventing muscle injury.

Another significant study by Bassini-Cameron et al. (2007) revealed that caffeine supplementation can increase the physiological response of athletes to exertion, creating a synergistic effect on white blood cell distribution and muscle damage markers. This effect is similar to that observed with the administration of natural antioxidants, which may intensify muscle recovery and protect the body from cellular damage.

Taking into account this information, this study focuses on the effects of natural SOD supplementation on the muscular and hepatic enzymatic profile of elite athletes. It aims at analyzing how natural SOD may influence key biochemical parameters, such as oxidative stress markers and muscle damage indicators, with the goal of improving performance and accelerating post-exercise recovery processes.

Hypothesis

The hypothesis of the study is that the administration of natural SOD will significantly reduce oxidative stress and modulate the muscular and hepatic enzymatic profile of elite athletes. The natural SOD enhances muscle recovery, reduces muscle damage and supports hepatic metabolic functions, as proved by the comparison with the control group that did not receive antioxidant supplementation.

The aim of the study is to evaluate the impact of natural SOD administration on the reduction of oxidative stress and the modulation of the muscular and hepatic enzymatic profile in elite athletes.

Material and methods

Research protocol

a) Period and place of the research

The retrospective clinical study was carried out in two distinct periods. Each period included two biochemical and functional evaluation tests: test I – before the administration of treatment and test II – after the administration of treatment. The male groups (MG) were evaluated during June and September 2021, while the female groups (FG) were evaluated during January and May 2022. The treatment consisted of administering the natural antioxidant SOD for a duration of 2.5 to 3 months, with a dosage of 2 ampoules per day for 60 days.

b) Subjects and groups

The subjects included in the study were high-performance athletes practicing football (average age, weight, height and BMI), totaling 38 male athletes – of which 26 received treatment (19.81 years, 73.07 kg, 179.64 cm and 22.6 kg/m²), while 12 did not receive treatment (16.83 years, 71.1 kg, 176.25 cm and 22.8 kg/m²). The female group consisted of 20 athletes, of which 16 received treatment (27 years, 61.73 kg and 21.8 kg/m²) while 4 did not follow treatment (24.5 years, 55.15 kg, 165.25 cm and 21.5 kg/m²).

Parental informed consent was obtained and signed in accordance with the Declaration of Helsinki before starting the research. The study was approved by the Ethics Committee for Scientific Research of the University of Pitești (No. 3915/05.04.2022), currently the National University of Science and Technology "Politehnica" Bucharest, Pitești University Center. The confidentiality of data and their protection are managed by the "Chiajna Medical Center".

The study was conducted with financial support from the "Cantacuzino" National Institute for Research and Development in Medical-Military Sciences of Romania, which provided the natural SOD antioxidant treatment. Among the partners and members of the research team were: the "Chiajna Medical Center", the National University of Science and Technology "Politehnica" Bucharest - Pitești University Center, the Doctoral School of Sports Science and Physical Education, and the State University of Physical Education and Sport of Chișinău, Republic of Moldova (currently, the State University of Moldova – Institute of Physical Education and Sport).

c) Applied tests (used instruments)

The medical-sports evaluation of the high-performance athletes was carried out at the "Chiajna Medical Center", where anthropometric and body composition data were collected. Blood samples were taken at the Medical Bio-Medica International SRL laboratory (based on a collaboration agreement). The following three parameters were analyzed: TGO (U/L), TGP (U/L) and creatine kinase (CK, U/L).

d) Statistical processing

The statistical analysis was performed using KyPlot 6.0 software (KyensLab Inc). Descriptive data included the mean and interquartile range (IQR). The data followed a normal distribution (tested with the Shapiro-Wilk test). There were used Wilcoxon Rank Sum Test (Mann-Whitney U Test) for unpaired and paired data (U, Z, P), effect size r=Z/√N for unpaired data and r – Spearman, for paired data. The confidence level was set at 95%, with statistical significance considered at p value ≤ 0.05.

Results

The study highlighted significant differences between the treated and untreated athlete groups, both in terms of age and anthropometric characteristics. These differences may influence the physiological response to treatment. The results show the influence of anthropometric characteristics and age on the physiological response to treatment, given that age and body composition can impact the effects of antioxidant supplementation on high-performance athletes.

The monitoring of muscle and liver enzyme values was conducted using non-parametric statistical analyses to point out significant differences between and within the study groups. To compare values between independent groups at test I (before treatment) and test II (after treatment), the Wilcoxon Rank Sum Test (Mann-Whitney U Test) for unpaired data was applied. Results are presented in Tables I and II.

The Wilcoxon Signed Rank Test for paired data was used for analyzing the changes within each group between the two testing moments (test I and test II). This analysis was applied to both the control group (CG), which did not receive treatment (Table III), and the experimental group (EG), which received the treatment with the antioxidant SOD (Table IV).

Additionally, comparative analyses between genders were performed for both CG (Table V) and EG (Table VI) to highlight potential differences in the response to treatment depending on gender.

Table I shows the results of the comparative analysis between groups in terms of muscle and liver enzyme values at Test I (before the application of the treatment).

TGO (AST - U/L) recorded higher values in both experimental groups. For males (M), the difference between the median and IQR was 5.72 U/L and 0.89 U/L, respectively. Although the difference is not statistically significant ($p > 0.05$), there is a trend towards higher values in the experimental group, with a small statistical effect. For females (F), the median difference was 7.99 U/L and the IQR difference was 7.91 U/L. Although the differences are not statistically significant here either, the p-value is close to the significance threshold ($p < 0.05$) and the effect is moderate ($r > 0.3$). These elevated values may reflect liver impairment or a mild degree of muscular stress before the application of the antioxidant treatment.

TGP (ALT - U/L) also showed increased differences in the experimental groups. For males (M), the median difference was 3.72 U/L and the IQR was 6.37 U/L, without statistical significance and with a small effect. For females (F), the values were higher in the experimental group, with a median difference of 7.07 U/L and an IQR difference of 9.675 U/L. Although the differences are not statistically significant, they suggest a possible state of hepatic stress before the administration of the treatment.

Creatine kinase (CK - U/L) revealed the largest differences between the control and experimental groups. For male (M), the median and IQR values were 334.47 U/L and 483.725 U/L, respectively, with a statistically significant difference ($p < 0.001$) and a moderate to large effect ($r > 0.5$). This indicates a high degree of muscular stress or a possible oxidative imbalance prior to treatment. For female (F), the difference between groups was also statistically significant, with a median value of 189.07 U/L and an IQR of 258.455 U/L; the effect was large ($r > 0.5$). These data support the hypothesis of intense muscular activity or increased oxidative stress among the female athletes in the experimental group.

Table II presents the results of monitoring muscle and liver enzyme values at Test II (after the administration of the treatment).

TGO (AST - U/L) had minimal differences between the male groups, with median values very close to 0.68 U/L and lower dispersion in the experimental group (IQR of 11.58). The difference was not statistically significant ($p = 0.7895$) and the effect was negligible ($r = 0.04$), indicating a similar enzymatic balance between groups. For female (F), the difference was statistically significant ($p = 0.0335$), with a higher median in the experimental group (7.57 U/L) and the effect was moderate ($r = 0.48$). This result may suggest a post-treatment reaction or intensified hepatic/ muscular activity in the female experimental group.

Table I

Results of the comparative analysis between groups regarding muscle and liver enzyme values at Test I.

Indicators	Gender	CG		EG		U	Z	P	r
		median	IQR	median	IQR				
TGO (U/L)	M	18.75	13.245	24.47	14.135	100	-1.76	0.0813	0.28
	F	18.07	2.37	26.06	10.28	12	-1.89	0.0654	0.42
TGP (U/L)	M	18.27	17.555	21.99	11.185	114	-1.32	0.1825	0.21
	F	13.22	3.535	20.29	13.21	19	-1.23	0.2375	0.27
CK (U/L)	M	153.18	84.715	487.65	568.44	36	-3.77***	0.0001	0.61
	F	154.23	35.905	343.3	294.36	0	-3.02**	0.0029	0.67

Notes: Male Control Group – MCG; Male Experimental Group – MEG; MCG, n= 12; FCG, n=4; MEG, n= 26; FEG, n=16; Wilcoxon Rank Sum Test (Mann-Whitney U Test) for Unpaired Data; √N38 = 6.164; √N20=4.472

Table II

Results of the comparative analysis between groups regarding muscle and liver enzyme values at Test II.

Indicators	Gender	CG		EG		U	Z	P	r
		median	IQR	median	IQR				
TGO (U/L)	M	21.43	22.13	22.11	10.55	165	0.28	0.7895	0.04
	F	18.06	3.91	25.63	6.6	9	-2.17*	0.0335	0.48
TGP (U/L)	M	18.75	16.03	18.86	8.02	174	0.56	0.5826	0.09
	F	14.32	5.06	20.78	7.29	16	-1.51	0.143	0.34
CK (U/L)	M	228.26	287.64	198.92	156.59	162	0.19	0.8629	0.03
	F	168.01	16.49	200.505	68.8	12	-1.89	0.0653	0.42

Notes: Male Control Group – MCG; Male Experimental Group – MEG; MCG, n= 12; FCG, n=4; MEG, n= 26; FEG, n=16; Wilcoxon Rank Sum Test (Mann-Whitney U Test) for Unpaired Data; √N38 = 6.164; √N20=4.472

Table III

Results of the intra-group comparative analysis of muscle and liver enzyme values in the control group.

Indicators	Gender	Test I		Test II		Z	P	r
		median	IQR	median	IQR			
TGO (U/L)	M	18.75	13.245	21.43	22.13	-3.06**	0.003	0.965
	F	18.07	2.37	18.06	3.91	-0.53	0.789	0.200
TGP (U/L)	M	18.27	17.555	18.75	16.03	-2.04*	0.045	0.930
	F	13.22	3.535	14.32	5.06	-1.83	0.100	1.000
CK (U/L)	M	153.18	84.715	228.26	287.64	-3.06**	0.003	0.930
	F	154.23	35.905	168.01	16.49	-1.83	0.100	0.800

Notes: Male Control Group (MCG), n= 12; Female Control Group (FCG), n=4; Wilcoxon Signed Rank Test for Paired Data.

Table IV

Results of intra-group comparative analysis of muscle and liver enzyme values in the experimental group.

Indicators	Gender	Test I		Test II		Z	P	r
		median	IQR	median	IQR			
TGO (U/L)	M	24.47	14.135	22.11	10.55	3.87***	0.0001	0.810
	F	26.06	8.805	25.63	6.6	0.10	0.938	0.894
TGP (U/L)	M	21.99	11.185	18.86	8.02	3.77***	0.0001	0.665
	F	20.29	13.21	20.78	7.29	-1.19	0.245	0.935
CK (U/L)	M	487.65	568.44	198.92	156.59	4.00***	0.0001	0.694
	F	343.3	284.36	200.505	68.8	3.46***	0.0006	0.706

Notes: Male Experimental Group (MEG), n= 26; Female Experimental Group (FEG), n=16; Wilcoxon Signed Rank Test for Paired Data

TGP (ALT - U/L) showed no significant differences for males ($p = 0.5826$), with nearly identical median values between groups (18.75 U/L in CG and 18.86 U/L in EG) and lower dispersion in the experimental group. The effect was very small ($r = 0.09$), indicating hepatic stability post-treatment. For females, although the values were higher in EG (0.11 U/L), the difference did not reach statistical significance ($p > 0.05$), but the effect was moderate ($r = 0.34$), which might reflect an individual metabolic response after the antioxidant administration.

Creatine kinase (CK - U/L) presented a decrease in median values in the male experimental group by 29.34 U/L and IQR of 131.05 U/L, but with no statistical significance ($p > 0.05$) and a very small effect ($r = 0.03$). This may prove a reduction in muscle stress after the treatment. For females, the values were slightly higher in EG by 32.5 U/L and IQR of 52.31 U/L, with a difference close to statistical significance ($p > 0.05$) and a moderate effect ($r = 0.42$), suggesting possible increased muscular activity or an individual response to treatment.

Table III shows the results of the intra-group comparative analysis of muscle and liver enzyme values in the control group (CG).

For TGO (AST - U/L), a significant increase was observed in the male control group (MCG), with median values rising by 2.68 U/L and an IQR of 8.885 U/L. This difference was statistically significant ($p < 0.01$), with a very large effect size ($r = 0.965$). This increase may be attributed to possible intense physical activity during the monitoring period or other environmental factors, in the absence of antioxidant treatment. For females (F), the values remained virtually unchanged between the two test moments, with a non-significant difference ($p > 0.05$) and a very small effect size ($r = 0.200$), revealing hepatic stability in the absence of treatment.

For TGP (ALT - U/L), a slight increase was observed in males, from a median value of 0.48 U/L and an IQR of 1.525 U/L, with a statistically significant difference (p

Table V

The results of the comparative analysis between male and female genders for muscle and liver enzyme values in the control group.

Indicators	Tests	Male		Female		U	Z	P	r
		median	IQR	median	IQR				
TGO (U/L)	Test I	18.75	13.245	18.07	2.37	28.5	0.55	0.627	0.138
	Test II	21.43	22.13	18.06	3.91	30	0.73	0.505	0.183
TGP (U/L)	Test I	18.27	17.555	13.22	3.535	32	0.97	0.363	0.242
	Test II	18.75	16.03	14.32	5.06	36	1.46	0.163	0.365
CK (U/L)	Test I	153.18	84.715	154.23	35.905	24	0.00	1.00	0.000
	Test II	228.26	287.64	168.01	16.49	40	1.94	0.060	0.485

Notes: Male, n= 12; Female, n=4; Wilcoxon Rank Sum Test (Mann-Whitney U Test) for Unpaired Data; √N16=4.00

Table VI

Results of the comparative analysis between male and female gender regarding muscle and liver enzyme values in the experimental group.

Indicators	Tests	Male		Female		U	Z	P	r
		median	IQR	median	IQR				
TGO (U/L)	Test I	24.47	14.135	26.06	8.805	213.5	0.14	0.897	0.022
	Test II	22.11	10.55	25.63	6.6	151	-1.48	0.143	-0.228
TGP (U/L)	Test I	21.99	11.185	20.29	13.21	266.5	1.52	0.133	0.235
	Test II	18.86	8.02	20.78	7.29	186	-0.57	0.578	-0.088
CK (U/L)	Test I	487.65	568.44	343.3	284.36	253	1.17	0.249	0.180
	Test II	198.92	156.59	200.505	68.8	237	0.75	0.460	0.116

Notes: Male, n= 26; Female, n=16; Wilcoxon Rank Sum Test (Mann-Whitney U Test) for Unpaired Data; √N42=6.48

< 0.05) and a large effect size (r = 0.930). This change, although numerically small, indicates enzyme variability that may be influenced by physical effort. For females (F), values increased by 1.1 U/L with an IQR of 1.525 U/L, but the difference was not statistically significant (p > 0.05), despite a maximum effect size (r = 1.000), likely due to the small sample size (n = 4).

For creatine kinase (CK - U/L), a significant increase was observed in the male control group, with a rise of 75.08 U/L and an IQR of 202.925 U/L, with p < 0.01 and a large effect size (r = 0.930). This suggests increased muscular stress between the two test moments, in the absence of antioxidant support. For females, CK values increased by 13.78 U/L with a decrease in IQR of 19.415 U/L, but the difference was not statistically significant (p > 0.05), with a moderate effect size (r = 0.800).

Table IV presents the results of the intra-group comparative analysis of muscle and liver enzyme values in the experimental group (GE).

In the case of TGO (AST - U/L), a slight decrease by 2.36 U/L and an IQR decrease by 3.585 U/L were observed in males (M). The difference was statistically significant (p < 0.001), with a very large effect size (r = 0.810), suggesting a possible hepatic protective effect as a result of the treatment. For females (F), the values decreased very slightly by 0.43 U/L, but the difference was not statistically significant (p > 0.05), with a very small effect size (r = 0.894), indicating hepatic stability with no important changes after the intervention.

As for TGP (ALT - U/L), a statistically significant decrease was observed in males, with a reduction of 3.13 U/L and IQR of 3.165 U/L, with p < 0.001 and a large effect size (r = 0.665). This decrease supports the hypothesis of a possible beneficial effect of the treatment on liver function. For females (F), the values increased slightly by 0.49 U/L, and the IQR increased by 5.92 U/L, but the difference was not significant (p > 0.05), with a reduced effect size (r = 0.935), indicating minor variability with no evident clinical significance.

For creatine kinase (CK - U/L), a statistically significant decrease was observed in the male experimental group (M), from a median of 288.73 U/L and an IQR of 411.85 U/L, with p < 0.001 and a strong effect size (r = 0.694). For females (F), values decreased by 142.795 U/L and the IQR decreased by 215.56 U/L, also with a statistically significant difference (p < 0.001) and a large effect size (r = 0.706). These results indicate a significant reduction in muscle stress post-treatment, suggesting the effectiveness of the intervention (possibly antioxidant) in reducing muscular strain or inflammatory processes.

Table V shows the results of the comparative analysis between male and female muscle and liver enzyme values in the control group (CG).

TGO (AST - U/L)

In Test I, males had higher values by 0.68 U/L at the median and IQR of 10.875 U/L. No significant differences were noticed between groups (p > 0.05), with a small effect (r = 0.138), pointing out a similar TGO value between males and females before treatment.

In Test II, the median for males was higher by 3.37 U/L and IQR of 18.22 U/L. The difference was not statistically significant (p > 0.05) and the effect was also small (r = 0.183). This indicates stability in the TGO values for both control groups, with no substantial changes after treatment.

TGP (ALT - U/L)

Test I: the median for males was higher by 5.05 U/L and IQR by 14.02 U/L. The differences between groups were not significant ($p > 0.05$) and the effect was small ($r = 0.242$).

Test II: the average values for Males Control Group (MCG) were 18.75 U/L (IQR = 16.03) and for Females Control Group (FCG) 14.32 U/L (IQR = 5.06). No significant differences were observed ($p = 0.163$) and the effect remained small ($r = 0.365$). This shows a similar progression of TGP in both groups.

Creatine Kinase (CK - U/L)

Test I: males had a lower median by 1.05 U/L and a higher IQR by 48.81 U/L, with a statistically insignificant difference ($p > 0.05$), revealing similar CK values in both groups before treatment.

Test II: after treatment, the median for males was higher by 60.25 U/L and IQR by 271.15 U/L. Although no significant differences were found ($p > 0.05$), there is a tendency for CK values to be higher in males after treatment, suggesting possible greater muscular activity or different recovery between males and females, with a moderate effect ($r = 0.485$).

Table VI presents the results of the comparative analysis between male and female in terms of muscle and liver enzyme values in the experimental group (EG).

TGO (AST – U/L)

Test I: the median for males was lower by 1.59 U/L and IQR higher by 5.33 U/L. No significant differences were observed between groups ($p > 0.05$) and the effect was very small ($r = 0.022$). These values suggest similarity between males and females before treatment regarding TGO levels.

Test II: after treatment, the median for males was lower by 3.52 U/L and IQR higher by 3.95 U/L. The differences were not significant ($p > 0.05$); the effect was negative and very small ($r = -0.228$). This suggests a trend towards higher TGO values in the female group, but without significant changes between groups.

TGP (ALT – U/L)

Test I: the median for males was higher by 1.7 U/L and IQR lower by 2.025 U/L. The differences were not significant ($p > 0.05$) and the effect was small ($r = 0.235$), indicating a similar TGP value before treatment.

Test II: the median for the male experimental group (MEG) was lower by 1.92 U/L and IQR higher by 0.73 U/L. The differences were not statistically significant ($p > 0.05$) and the effect was small ($r = -0.088$), highlighting stability in TGP values after treatment, with no significant differences between males and females.

Creatine Kinase (CK – U/L)

Test I: the median for males was higher by 144.35 U/L and IQR by 284.08 U/L. The difference between groups was not statistically significant ($p > 0.05$), with a small effect ($r = 0.180$). This shows that no significant differences were noticed between males and females before treatment, as for creatine kinase levels.

Test II: the median for males was lower by 1.585 U/L and IQR higher by 87.79 U/L. Differences were not statistically significant ($p > 0.05$) and the effect was also small ($r = 0.116$), indicating similar stability in CK levels between the two groups after treatment.

Discussion

The results obtained after the administration of the natural antioxidant SOD demonstrate a significant improvement in biochemical and functional parameters post-treatment, both in male and female groups, suggesting a beneficial effect of antioxidant supplementation on the endurance capacity and oxidative status of the elite athletes.

In the comparative analysis of the anthropometric data of high-performance athletes, it is essential to consider factors that may influence physical parameters and physiological responses to antioxidant treatments, including gender differences and the impact of antioxidant supplementation. Recent studies highlight the effectiveness of antioxidant supplementation, including SOD, in optimizing recovery and performance in athletes by reducing muscle damage and enhancing antioxidant function. These aspects are supported by the research of Gravina et al. (2012), Aguinaga-Ontoso et al. (2023), García-Cardona et al. (2021) and Petri et al. (2024), who demonstrate the positive impact of these interventions on physiological responses and adaptations to intense training.

The values of muscle and liver enzymes recorded in Test I of Table I emphasize the presence of initial differences between the experimental and control groups, particularly in relation to creatine kinase (CK) activity. This fact indicates a possible elevated level of muscle stress or pre-existing oxidative imbalance in the subjects of the experimental groups, justifying the need for antioxidant treatment intervention. The analysis of muscle and liver enzyme values in Test II of Table II shows a relatively stable enzymatic profile post-treatment, especially in males, where the differences between the experimental and control groups are minimal and statistically insignificant. In contrast, in females, slightly more pronounced metabolic responses are observed in the experimental group, with moderate intensity effects, suggesting possible individual variability in response to antioxidant administration.

The studies of Toro-Román et al. (2023) and Souglis et al. (2023) identify significant gender differences in heavy metal concentrations and metabolic responses to oxidative stress and inflammation, emphasizing the need for a personalized approach to optimizing the performance and recovery of athletes, particularly in the context of intense training and antioxidant treatments.

The intra-group comparative analysis presented in Table III within the control group (CG) highlights a significant increase in muscle and liver enzyme values in male subjects, particularly for TGO and CK, indicating a potential accumulation of physiological stress and muscular stress in the absence of antioxidant intervention. Conversely, female subjects exhibit a relatively stable enzymatic profile, with statistically insignificant variations, although moderate to large effects may be influenced by the small sample size.

The results of the intra-group analysis in the experimental group (EG) from Table IV reveal significant decreases in the values of TGO, TGP and CK in male subjects, supporting the effectiveness of the applied treatment, possibly with an antioxidant role, in the diminution of the hepatic and muscular stress. In females, changes were insignificant for

hepatic parameters, but the substantial reduction in CK values confirms a positive physiological response of the musculature to the intervention, confirming a beneficial effect on the neuromuscular system, particularly. Recent studies underscore the importance of managing muscle damage, inflammation and oxidative stress in team sports, indicating that intense training and multiple competitions can trigger significant physiological responses. Interventions such as supplementation with astaxanthin (Baralic et al., 2015), the consumption of polyphenols (Sanchez Diaz et al., 2022) and the management of antioxidant-rich diets (Zare et al., 2023) have been shown to reduce these effects and improve the recovery of athletes, as demonstrated by Mohr et al. (2016) in the context of football.

The comparative analysis between genders in the control group in Table V shows no significant statistical differences in liver and muscle enzyme values (TGO, TGP, CK) between males and females, both in Test I and Test II, revealing a relatively uniform enzymatic progression in the absence of treatment. However, the slightly higher CK values in males in Test II, accompanied by a moderate effect, may indicate different muscular stress or recovery capacities specific to males, an aspect worthy of exploration in future studies.

The comparative analysis between genders in the experimental group in Table VI proves the absence of significant statistical differences between males and females, both before and after treatment, regarding the values of TGO, TGP and CK, with very small or small effects in all cases. These results suggest that the applied intervention had a similar impact on both genders, with no notable variations in hepatic or muscular responses, confirming biological balance between sexes in the context of antioxidant treatment administration.

Recent studies emphasize the impact of dietary supplements and personalized interventions on athletes' performance and recovery, emphasizing their effectiveness in optimizing health and athletic outcomes, the influence of matches on muscle damage and physiological responses in amateur athletes (Dewangga et al., 2024) and gender differences in post-exercise inflammatory responses, which require adaptive recovery strategies (Stupka et al., 2000). The study conducted by Mitrotasios et al. (2021) highlights the effects of Small Sided Games (SSGs) training on the biochemical profile of soccer players, demonstrating their impact on changes in lipid, enzymatic and hormonal markers, which can help coaches optimize high-intensity training sessions to enhance player performance and health.

These findings contribute to a deeper understanding of the physiological mechanisms involved in athlete recovery and the development of more effective strategies to optimize performance in conditions of intense training and repeated competitions.

Conclusions

1. The administration of the natural antioxidant SOD led to significant improvements in biochemical and functional parameters post-treatment, confirming its effectiveness in reducing oxidative stress and supporting endurance capacity in high-performance athletes, regardless of gender.

2. The comparative analysis of anthropometric data reveals relevant differences between the male and female subgroups, influenced by age, weight and height, but without notable variations in body mass index, suggesting a balanced body composition between treated and untreated subjects.

3. The elevated CK values in the experimental groups prior to treatment indicate intense muscular stress, justifying the need for antioxidant intervention. The subsequent reduction in these values in Test II confirms a protective muscle effect of SOD, particularly in male athletes.

4. The absence of significant statistical differences between genders, both in the control group and the experimental group, suggests a similar physiological response to antioxidant treatment, reflecting an enzymatic balance between boys and girls and validating the applicability of the treatment in both categories of athletes.

Conflict of interest

The authors declare no conflict of interest.

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