

ORIGINAL RESEARCH

The effect of endurance training on cardiac and muscle damage in male football players

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Abstract

This study aimed to investigate the effects of a six-week endurance training program on heart and muscle damage in football players. Twenty male football players from the same team voluntarily participated in the study. Participants were randomly divided into two groups: the experimental group (n = 10; mean age 22 ± 2.21 years) and the control group (n = 10; mean age 22.3 ± 1.76 years). On the first day of the study, 3 mL of venous blood samples were collected into ethylenediaminetetraacetic acid tubes by a specialist to measure N-Terminal Pro-B-Type Natriuretic Peptide for cardiac damage and Creatine Kinase (CK) for muscle damage. On the second day of the study, the Yo-Yo Intermittent Recovery Test Level-1 (YIRT1) was conducted to assess the players' maximum oxygen consumption (VO₂max) and maximum aerobic speed (MAS) values. After six weeks of endurance training, the procedures above were repeated. A statistically significant difference between the pre-test and post-test data of CK, NT-ProBNP and VO₂max in the experimental group ($p < 0.01$) was observed. No statistically significant difference was observed between the pre and post-test data in the control group ($p > 0.05$). Additionally, based on the analysis of variance repeated measured analysis, a statistically significant difference was observed between the experimental and control groups regarding the time and time × group variables for NT-ProBNP and VO₂max parameters ($p < 0.01$). However, no statistically significant difference was observed for CK levels ($p > 0.05$). A statistically significant difference was observed in CK levels in terms of the group variable ($p < 0.01$), while no statistically significant difference was observed in NT-ProBNP and VO₂max levels ($p > 0.05$). Ultimately, our study revealed that endurance training positively affects heart and muscle damage.

Keywords

Brain natriuretic peptide; Creatin kinase; Maximal aerobic speed

1. Introduction

The need for physical exertion in contemporary football is progressively rising [1]. Regarding the evolution of football, critical factors for enhancing performance include linear speed, explosive movements and high-intensity maneuvers such as jumping, acceleration, deceleration, change of direction (COD) and functional mobility [2]. Various factors can influence the performance component in football. One of the most essential aspects, endurance performance, is recognized to affect match performance greatly [3]. As an illustration, during a 90-minute football match, a player often covers a distance of 10 to 12 kilometers while maintaining an average effort level up to the anaerobic threshold of 80% to 90% of their maximal heart rate. They can perform 150–250 high-intensity sprints of 15–20 meters and a variety of explosive movements [4]. The sprints and actions significantly impact the performance of individual players and the team, potentially changing the result of a match [5]. As a result, strength and

conditioning experts are focusing more on physical fitness development alongside ever-changing strategies and methods [6].

Endurance training improves exercise performance while increasing oxygen delivery, substrate metabolism and mitochondrial mass in skeletal muscle [7]. In addition to endurance training, skeletal muscle experiences considerable stress, leading to exercise-induced muscle damage [8]. This disruption in muscle fiber integrity is frequently linked to muscle damage, which includes delayed onset muscle soreness (DOMS), leakage of intramuscular enzymes such as creatine kinase (CK) into circulation, and impaired muscle function. CK levels are known to rise with both high-intensity sustained exercise and eccentric muscular contractions [9]. Felismino *et al.* [10] observed that the presence of CK in the bloodstream indicates muscle damage, providing valuable insights into muscle integrity and the athlete's performance. Toutsoka also observed that levels of the creatine kinase enzyme increase following endurance training [11]. Mariana *et al.* [12] found in a

distinct study that participating in low-intensity physical activity results in a significant reduction in creatine kinase levels. Furthermore, studies have demonstrated that participating in vigorous exercise, namely at intensities ranging from 70% to 90% of the maximum repetition, can result in a decrease in serum creatine kinase levels.

Endurance training is a highly effective approach to enhance various physiological parameters essential to performance, such as cardiometabolic function, oxygen uptake kinetics, anaerobic power and capacity, ion transport capacity and muscle fatigue resistance [13]. Endurance training enhances cardiovascular factors, including cardiac output, blood flow rate and arterial distensibility, in football players [14]. These developments improve the oxygen uptake capacity of an athlete. Consequently, football players' aerobic capacity is enhanced, improving their ability to sustain high-intensity exercise [15]. After engaging in consistent endurance training, the heart undergoes structural and functional changes [16]. Changes occurring in athletes as a response to increased and repeated hemodynamic stress due to chronically applied training are called athlete's heart. These modifications involve myocardial hypertrophy to enhance contraction and chamber dilation to accommodate a higher blood volume. Functionally, trained individuals exhibit higher ejection fractions to meet the increased oxygen demand during physical exertion and lower resting heart rates [17]. Brain natriuretic peptide (type B) (BNP) and its more stable inert form, the (N-terminal) amino moiety (NT-ProBNP), are widely recognized as the most reliable method for determining cardiac muscle tension [18]. N-Terminal Pro b-type natriuretic peptide (NT-ProBNP), a member of the natriuretic peptide family, is mainly secreted by the ventricles and, to a lesser extent, by the atria [19]. NT-ProBNP, a marker for identifying cardiovascular disease, can be tested using immunological methods. However, it is essential to note that NT-ProBNP levels increase when there is left ventricular hypertrophy [20].

BNP is secreted in response to the stimulation of receptors in ventricular muscle cells, resulting from stretching or damage of the ventricles. BNP levels are a marker of left ventricular ejection fraction and cardiovascular endurance [21]. The ventricles are the primary source of BNP, making it more sensitive and specific in detecting problems in the ventricles than other natriuretic peptides. As a result, rapid detection and treatment of BNP secretion minimizes heart health risks and plays a major part in decreasing mortality and morbidity rates among persons with this illness [22].

This research is anticipated to offer substantial input to understanding the long-term effects of endurance training on football players. The study's findings could provide valuable insights into optimizing training programs to boost athletes' cardiovascular and muscular health without adverse effects. Furthermore, monitoring BNP and CK levels can be useful for tracking athletes' heart and muscle health and early detection of potential health issues. The synchronized operation of the heart and muscles is expected to result in elevated levels of physical performance. When the heart adequately supplies oxygen and nutrients to the muscles, the muscles function more efficiently and exhibit increased endurance. Regularly monitoring CK levels is vital for strengthening muscles and im-

proving endurance. Endurance training induces micro-damage to the muscles, resulting in an initial elevation of CK levels. Consistent and prolonged endurance training can enhance muscle adaptation, contributing to a long-term decrease in CK levels. Hence, the planning and implementation of endurance training are vital for maintaining muscle health and managing CK levels. By implementing effective training strategies and employing appropriate recovery procedures, it is possible to manage muscle damage and enhance endurance effectively.

As previously mentioned, NT-ProBNP is a marker used to detect heart damage. While very few researches explore BNP levels in athletes [23, 24], there is little or no evidence of studies examining NT-ProBNP levels. Our study examines the use of monitoring BNP levels after endurance training to evaluate athletes' performance and the effectiveness of training programs. We hypothesize that changes in BNP levels and improvements in endurance performance (measured by increased $VO_2\max$) may indicate enhanced performance. Additionally, measuring BNP levels throughout this process could be useful for assessing heart health. Tracking BNP levels alongside athletes' performance could be regarded as an indicator of heart health. Nevertheless, numerous studies have examined the levels of BNP in persons with cardiac disease who undergo a chronic exercise program [25, 26]. The impact of endurance training on heart and muscle damage in football players is not fully understood. Understanding the long-term effects of endurance training on both cardiovascular and muscular health is imminent for maximizing athletes' training programs and safeguarding their overall well-being. The objectives of this study are as follows: (1) To evaluate BNP levels in athletes as a result of induced cardiac damage following applied endurance training, (2) to assess creatine kinase (CK) levels in athletes as a result of induced muscle damage following applied chronic training. Our hypothesis posits that the training performed by football players will reduce BNP and CK levels while simultaneously increasing their maximal oxygen uptake ($VO_2\max$) capacities.

2. Materials and methods

2.1 Study participants

Twenty male amateur football players who actively played football volunteered to participate in the study. The study was conducted in the interim period after the end of the first competition period. The participants were randomly divided into 2 groups: the experimental group ($n = 10$; mean age, 22 ± 2.21 years) and the control group ($n = 10$; mean age, 22.3 ± 1.76 years) (Table 1). While weekly routine training (3 days per week) continued in the experimental and control groups, endurance training using the maximal aerobic speed method (MAS) was also performed in the experimental group. Athletes were notified about the study, and their participation was voluntary.

2.2 Methods

The pre and post-test control group model was employed in this research. On the first day of the study, blood samples (venous blood samples; 3 mL) were taken from the athletes

TABLE 1. Demographic characteristics of participants.

	Age (yr)	Height (cm)	Weight (kg)
EG			
Mean	22.00	180.30	73.20
SD	2.210	7.930	12.76
CG			
Mean	22.30	174.90	71.70
SD	1.76	5.40	10.09

EG: experimental group; CG: control group; SD: standard deviation.

by the specialist physician and placed into tubes containing EDTA. On the second day of the study, the Yo-Yo Intermittent Recovery Test Level 1 (YIRT1) was applied to determine the MAS values of the athletes. After 6 weeks of endurance training, the procedure was repeated. The working procedure is illustrated in Fig. 1.

2.3 Maximal aerobic speed training program

The maximal aerobic speed training method (MAS) was used in our study. Athletes were administered a 6-week training program ranging from 70% to 100% intensity [27]. The training content is shown in Table 2. The athletes' MAS value was calculated using the formula "MAS = final speed (km/h) \times 1.34 - 2.86" based on their performance in the Yo-Yo test [28]. Participants' VO_2 max was calculated using the following formula: VO_2 max (mL/kg/min) = 0.0084 \times Final Distance (meter) + 36.4 [29].

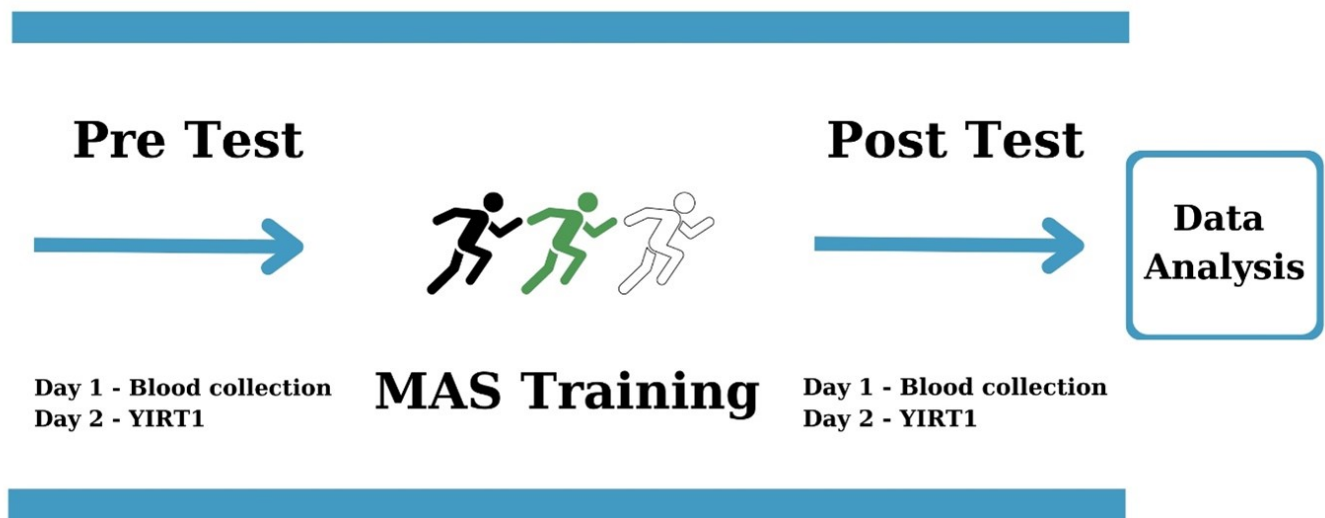
2.4 Data sources and data collection method

Fig. 2 illustrates the Yo-Yo test. The route spans from point A to point B. During the run, a beep will sound as you reach point B from A, prompting you to turn around and sprint back to point A. When you reach point A, the beep sounds again, and you jog from A to C and then wait until the beep sounds again at point A. While this is ongoing, the Yo-Yo test is conducted simultaneously. The running speed increases according to the test protocol. If an athlete misses the signal the first time he reaches point A, a faulty beep will sound. Each time the athlete arrives at A, the test distance is marked and recorded on the Yo-Yo test form.

The Yo-Yo test should be conducted on consistent ground and in-weather conditions. Athletes are required to wear shoes suitable for the test ground. The initial speed of the test is 10 km/h. The running speed increases by 0.5 km/h every 40 meters [30].

2.5 Biochemical analyze

A physician specialist collected venous blood samples (3 mL) from the athletes into EDTA tubes. The blood samples were gently inverted 3–5 times to prevent hemolysis. After 20 minutes, they were centrifuged at 3500 rpm for 5 minutes. The serum at the top part of the tube was transferred into Eppendorf tubes using a pipette. The Eppendorf tubes were stored at -80°C until they were taken out for analysis. CK and NT-ProBNP parameters were examined in blood serum samples [31]. CK analysis was conducted on a Beckman Coulter autoanalyzer (autoanalyzer, Beckman coulter, Brea, CA, USA) using a Beckman Coulter CK kit, employing the kinetic ultraviolet method. The NT-ProBNP analysis was performed on a Snibe-Maglumi X8 (Immunoassay analyzer, Snibe Diagnostic, Shenzhen, Guangdong, China) fully automated chemiluminescence immunoassay (CLIA) analyzer using a Maglumi NT-ProBNP kit, utilizing the chemiluminescence method.



Notes. YIRT1 = Yo-Yo Intermittent Recovery Test-1; MAS = Maximal Aerobic Speed.

FIGURE 1. Experimental design. A scheme of blood collection, YIRT1 and subsequent analyses performed.

TABLE 2. MAS training program.

	1st Week	2nd Week	3rd Week	4th Week	5th Week	6th Week
Intensity of Training	70% MAS	75% MAS	80% MAS	85% MAS	90–95% MAS	100% MAS
Training Frequency (d/wk)	3 d/wk	3 d/wk	3 d/wk	3 d/wk	3 d/wk	3 d/wk
Training Time	4 × 6 min	4 × 6 min	6 × 4 min	6 × 4 min	4 × 4 min	4 × 4 min
Rest Time Between Sets	3 min	3 min	2 min	2 min	3 min	3 min

MAS: maximal aerobic speed; d/wk: day/week; min: minute.

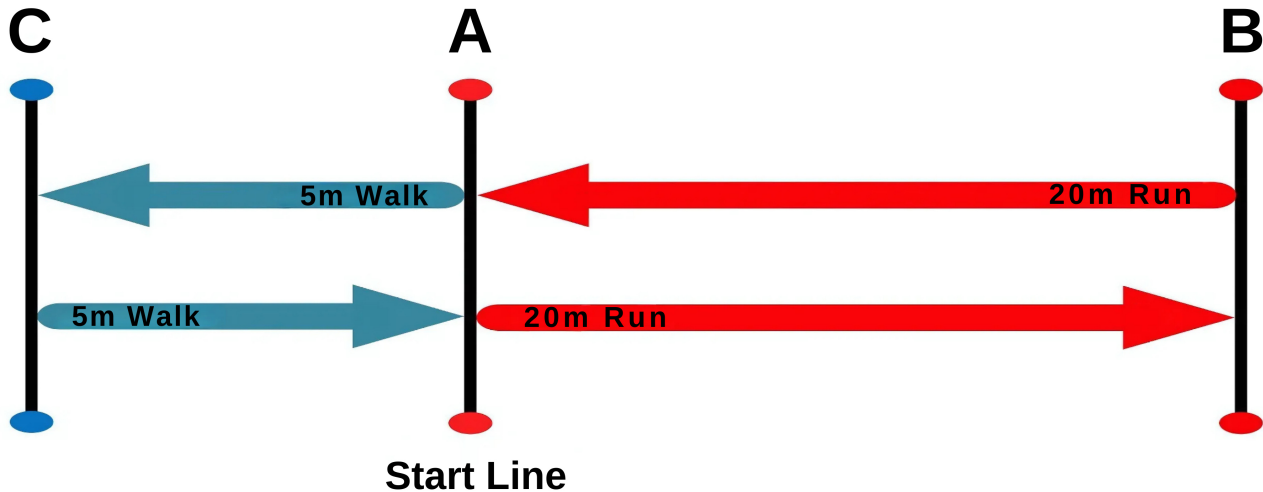


FIGURE 2. Yo-Yo intermittent recovery test level 1.

2.6 Data analysis

Following confirmation of data distribution normality through the Shapiro-Wilk test, repeated measures ANOVA was employed to evaluate the training impact on VO_2max , CK and NT-ProBNP values among football players following a 6-week MAS training program. Intra-group differences were examined using paired samples *t*-tests, while independent samples *t*-test was used to evaluate pre-test variations between groups. The study's effect size was quantified using Cohen's *d* analysis.

3. Results

The study results indicate no statistically significant difference in the pre-test NT-ProBNP levels between the experimental and control groups ($p > 0.05$). However, a statistically significant difference was observed between the post-test NT-ProBNP levels ($p < 0.05$).

When examining the results of the repeated measures ANOVA, statistically significant differences were detected between the pre-test and post-test values for the experimental and control groups in VO_2max and NT-ProBNP when compared by time and time × group variables. However, no statistically significant difference was found in CK levels. Regarding the group variable, statistically significant differences were found among CK levels ($p < 0.05$). However, no statistically significant differences were detected between NT-ProBNP and VO_2max levels ($p > 0.05$) (Table 3).

There was no statistically significant difference between the pre-post test data for the control group ($p > 0.05$). However, a

statistically significant difference was found between the pre-test and post-test parameters for the experimental group ($p < 0.05$). Upon reviewing the study findings, it was observed that after 6 weeks of training, there was a decrease in CK and NT-ProBNP levels in the experimental group athletes, while an increase was noted in VO_2max values (Table 4).

4. Discussion

The primary aim of this study is to examine the impact of 6 weeks of endurance training on heart and muscle damage in football players. The study results revealed a statistically significant difference in the pre and post-test NT-ProBNP values of the experimental group ($p < 0.05$). Furthermore, the pre-test NT-ProBNP values of the experimental and control groups showed no statistically significant difference ($p > 0.05$). In contrast, a statistically significant difference was found between the post-test NT-ProBNP values ($p < 0.05$).

Endurance training enhances the mechanical robustness of the heart by improving cardiac output and myocardial endurance [32]. NT-ProBNP and BNP are the primary markers of cardiac damage [33]. BNP is generally a biomarker for diagnosing heart failure [34]. BNP is secreted by the ventricles in response to ventricular volume expansion and pressure overload [35]. A comprehensive literature analysis revealed a lack of long-term research investigating the levels of BNP in football players. Several studies have established that BNP primarily indicates heart damage in athletes during intense physical activity [36, 37]. It could be argued that measuring BNP levels after intense exercise is useful for evaluating heart

TABLE 3. Repeated measured ANOVA results of the participants.

Group	Pre-test	Post-test	Time	Time × group	Group
VO ₂ max (mL/kg ⁻¹ /min ⁻¹)					
EG	45.2 (±2.90)	49.753 (±3.39)	<0.001**	<0.001**	0.140
CG	45.9 (±2.92)	45.270 (±1.99)			
CK (U/L)					
EG	288.3 (±135.4)	190.2 (±73.91)	0.262	0.279	0.007**
CG	387.9 (±172.1)	386.1 (±172.1)			
NT-ProBNP (pg/mL)					
EG	672.6 (±142.8)	482.4 (±70.32)	0.009**	<0.001**	0.953
CG	553.3 (±25.32)	598.1 (±68.6)			

Note: ***p* < 0.01; EG: Experimental Group; CG: Control Group; VO₂max: Maximum Oxygen Uptake; NT-ProBNP: N-Terminal Pro b-type Natriuretic Peptide; CK: Creatine Kinase.

TABLE 4. Comparison of CK, NT-ProBNP and VO₂max parameters of the groups.

Group	Variable	N	Mean	SD	<i>t</i>	<i>p</i>	Cohen's <i>d</i>
CG							
	CK Pre Test (U/L)	10	387.92	172.16	0.23	0.982	
	CK Post-Test (U/L)	10	386.10	173.44			
	NT-ProBNP Pre-Test (pg/mL)	10	553.36	25.32	-1.98	0.080	
	NT-ProBNP Post-Test (pg/mL)	10	598.11	68.66			
	VO ₂ max Pre-Test (mL/kg ⁻¹ /min ⁻¹)	10	45.90	2.92	1.45	0.181	
	VO ₂ max Post-Test (mL/kg ⁻¹ /min ⁻¹)	10	45.27	2.00			
EG							
	CK Pre-Test (U/L)	10	288.30	135.46	3.74	0.005**	0.62
	CK Post-Test (U/L)	10	190.20	73.92			
	NT-ProBNP Pre-Test (pg/mL)	10	590.30	51.41	6.59	<0.001**	1.00
	NT-ProBNP Post-Test (pg/mL)	10	534.40	59.11			
	VO ₂ max Pre-Test (mL/kg ⁻¹ /min ⁻¹)	10	45.27	2.91	-14.71	<0.001**	1.42
	VO ₂ max Post-Test (mL/kg ⁻¹ /min ⁻¹)	10	49.75	3.39			

Note: ***p* < 0.01; EG: Experimental Group; CG: Control Group; VO₂max: Maximum Oxygen Uptake; NT-ProBNP: N-Terminal Pro b-type Natriuretic Peptide; CK: Creatine Kinase; SD: standard deviation.

health and tracking athlete performance. Utilizing markers like BNP to assess the impact of training on athletes might also aid in monitoring the recovery process after exercise. BNP levels may influence performance, particularly in athletes who undergo rigorous training or participate in lengthy races like marathons. Nonetheless, certain studies have investigated the levels of BNP and the effects of a long-term exercise program on persons with cardiac disease [38–41]. Ahmed Elshazly *et al.* [42] conducted a study to assess the effects of low-intensity exercise training on serum C-Reactive Protein (CRP) and BNP levels, as well as cardiac function indices, in patients with heart failure with reduced ejection fraction (HFrEF). The findings demonstrated that exercise significantly reduced BNP levels but did not directly affect CRP levels.

Nascimento *et al.* [43] performed a meta-analysis investigating the effects of endurance training on myocardial stress and inflammation in patients with heart failure. They reported that endurance training positively affected natriuretic peptides

and inflammatory biomarkers. It also leads to a reduction in BNP levels among participants. Similar to previous studies, Edwards and O'Driscoll implemented the high-intensity interval training (HIIT) method three days per week for twelve weeks in patients diagnosed with heart failure. According to the study, the BNP levels of the participants decreased [44]. The decrease in BNP levels following exercise suggests improved cardiac muscle function and less cardiac stress. This indicates that exercise has the potential to enhance cardiovascular health in persons diagnosed with heart disease and promote optimal functioning of the heart. It can also be linked to improved cardiovascular health and less cardiac strain. The decrease in BNP levels among athletes appears to be attributed to higher VO₂max and lower EF. Thus, it can be deduced that there is an inverse relationship between VO₂max and NT-ProBNP levels. In his work, Smart proposed that VO₂max responses could serve as a marker for BNP levels [45]. Passino *et al.* [46] found a negative correlation between BNP levels

and $VO_2\text{max}$. Our study found that although the $VO_2\text{max}$ of the athletes improved, the NT-ProBNP level decreased.

The extent to which athletes suffered muscle damage was also investigated in our study. The athletes' serum levels were analyzed at CK to assess the extent of muscle damage. CK levels generally increase during prolonged and vigorous activity and eccentric muscular contractions [47]. Kobaner [48], discovered that CK in the bloodstream is associated with muscle damage, indicating an athlete's muscle health and performance. Upon analyzing the findings of our study, we discovered a statistically significant difference in the pre- and post-test outcomes of the experimental group ($p < 0.05$). Likewise, it was found that the CK scores of the experimental group decreased in the post-test as compared to the control group. The training led to a marked decrease in the CK concentration of the experimental group. Consequently, it is hypothesized that, organisms acclimate to oxidative stress through training, leading to a drop in CK levels. The training program employed by athletes can be attributed to a decrease in CK levels.

5. Conclusions

Our study has shown that endurance training positively affects both heart and muscle damage. This study proposes a potential correlation between $VO_2\text{max}$ and NT-ProBNP. Hence, the correlation between these two variables might be explored in further investigations. Additionally, it is recommended that NT-ProBNP be used as a marker in acute and chronic studies involving footballers to evaluate their heart damage. Monitoring NT-ProBNP levels in footballers is considered to help discover potential heart problems early. Therefore, regular measurement of this biomarker in footballers is considered essential.

6. Strengths, limitations and future perspectives

This study examines the effects of endurance training on heart and muscle damage in football players. It is the first study to chronically investigate NT-ProBNP, a cardiac damage marker, in football players. The study's findings indicate that endurance training in football players reduces CK and NT-ProBNP levels and improves performance. However, some limitations should be considered. The players were amateur, and the sample size was small. Additionally, the players were not in the same conditions regarding nutrition, accommodation, sleep patterns, *etc.*, which are fundamental limitations. Considering these conditions, more research is needed to validate the application's variables. Furthermore, monitoring CK and NT-ProBNP levels in athletes is crucial for performance and tracking heart and muscle health. These biomarkers can help detect early signs of overtraining, muscle damage, and cardiac stress, enabling timely measures to protect the athletes' health and prevent potential injuries.

ABBREVIATIONS

BNP, brain natriuretic peptide; CK, creatine kinase; MAS, maximum aerobic speed; NT-ProBNP, N-Terminal pro b-type natriuretic peptide; $VO_2\text{max}$, maximum oxygen uptake; YIRT1, Yo-Yo intermittent recovery test level 1; EG, Experimental group; CG, Control group.

AVAILABILITY OF DATA AND MATERIALS

The data presented in this study are available on reasonable request from the corresponding author.

AUTHOR CONTRIBUTIONS

KG and KN—designed the research study; KG, KN and HA—performed the research; KN and HA—provided help and advice on critical review of the manuscript; HA—analyzed the data; ÖP—statistical analysis, writing-review & editing; KG—the manuscript. All authors contributed to editorial changes in the manuscript. All authors read and approved the final manuscript.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

This research was carried out following the ethical principles of the Declaration of Helsinki. The Ethics Committee of the Faculty of Medicine of Karamanoğlu Mehmetbey College (Decision No: 01-2023/25) approved the study. The confidential treatment and anonymity of the data was guaranteed according to the European Data Protection Law. Informed consent was obtained from all participants before starting the research.

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CONFLICT OF INTEREST

The authors declare no conflict of interest. Additionally, the decision to share data upon reasonable request is a collective decision made by all authors.

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