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Research Article

Effects of Beta-alanine Supplementation on Recovery and Performance Factors in Male Soccer Players

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Abstract

Background: There is growing interest in using dietary supplements to delay fatigue, accelerate recovery time, and improve the performance of athletes.

Objectives: In the present study, we investigated the effect of beta-alanine supplementation on the levels of some metabolic indexes related to recovery and the performance of soccer players.

Methods: Sixteen young soccer players [age: 20 ± 1.19 years, weight: 67.67 ± 8.9 kg, height: 176.38 ± 5.54 cm and body mass index (BMI): 21.76 ± 2.12 kg/m² completed this study. The subjects were randomly assigned into two groups. They were supplemented with 5g/day beta-alanine (BA) (BG, N = 8) or placebo (PG; N = 8; maltodextrin) for three weeks. Maximum oxygen consumption, explosive power, and recovery-related blood parameters (creatine kinase, lactate dehydrogenase, urea) were measured in two stages before and after supplementation.

Results: No significant change was observed in the serum creatine kinase level after the intervention (P = 0.061). Also, no significant difference was observed in serum levels of other blood factors, such as lactate dehydrogenase (P = 0.061) and urea (P = 0.061). Regarding performance factors, no significant changes were observed in aerobic power (P = 0.061) and explosive power (P = 0.061). **Conclusions:** Our results suggest that beta-alanine has no effect on reducing metabolic factors related to recovery and may improve the aerobic power of soccer players.

Keywords: Beta-alanine, Recovery, Metabolic Index, Aerobic Power, Soccer

1. Background

Today, the strategy of using effective and permissible dietary supplements to delay fatigue, accelerate recovery time, and improve athletes' performance is very common among coaches and athletes. Also, in modern football, because the nature of this sport is movements with different intensities and the pressure of the match is high, supplements are used to improve performance and recovery (1).

A professional player can cover distances between 10 and 13 km, which includes 1,500 to 3,000 meters of intense activity (moving > 18 km/h) during a match (2). Highintensity physical activity with limited opportunities for recovery can affect homeostasis (3). During a season, the chances of cramps are very high due to stressful training and games. As a result, there is a lot of emphasis on postexercise and competition recovery (4).

The physiological stress inflicted on soccer players is associated with some fatigue-related blood biomarkers, which are important biomarkers of a soccer player's physical effort after a training session and match (5), as previous research has shown that high levels of some of these biomarkers, such as creatine kinase, lactate dehydrogenase, and urea, after a soccer match or training up to 72 hours later, are associated with a decrease in strength and speed performance (6, 7).

Proper recovery immediately after the match can help improve an athlete's performance. Ergogenic dietary supplements can enhance energy production, regulate body composition, and improve performance. Beta-alanine is one of the supplements that has recently attracted much interest. This growing interest stems from recent research that has identified beta-alanine as the carnosine precursor (8), which, among other roles, appears to be an essential physiological buffer. It can easily absorb protons during contraction-induced acidosis (9). It has been well established that a daily intake of 1.6 to 6.4 grams of beta-alanine for 2 to 10 weeks can significantly increase muscle carnosine concentration (10, 11). Increasing muscle carnosine

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concentrations may improve athletic performance during exercise, leading to increased muscle acidity. It can also have potent antioxidant effects. Thus, beta-alanine has indirect and protective effects on the cell and modulates cell damage caused by intense sports activities (12).

2. Objectives

The current study sought to investigate the effect of beta-alanine supplementation on metabolic parameters related to recovery after an intense football game and whether it leads to accelerated recovery in the training session of the next match. Also, following the increase of muscle carnosine, an improvement in athletes' performance is expected.

3. Methods

3.1. Participants

Among the eligible players from the university soccer team, sixteen people who met the research inclusion criteria participated in this study [mean \pm SD; age: 20 \pm 1.19 years, weight: 67.67 ± 8.9 kg, height: 176.38 ± 5.54 cm and body mass index (BMI): $21.76 \pm 2.12 \text{ kg/m}^2$]. All participants, who were healthy and free of any disease, were selected and divided randomly and equally into the two groups of supplements (N = 8) and placebo (N = 8) (Figure 1). All participants had an average of four sessions per week of soccer training and abstained from taking betaalanine supplementation for at least the last six months or any other exercise supplement for at least the previous three months before beginning the study. Before the commencement of the trial, written informed consent was obtained from each participant after the study details were shared in depth with them.

3.2. Assessment of Exercise Performance

Yo-Yo and CMJT tests were used to check the aerobic and anaerobic performance of the subjects before the supplementation course. The exercise performance tests were conducted after the end of the match to equalize the pressure on the subjects and check the level of physical performance decline after a match (Figure 1).

3.2.1. Yo-Yo Intermittent Fitness Test

The Yo-Yo intermittent fitness test is a test of the ability to do repeated high-intensity exercise. Participants run back and forth on a twenty-meter course, with a ten-second active break after each 40 m, with speed increasing at set intervals until they cannot continue (13) (Figure 2). VO2 max, as an indicator of aerobic capacity, was obtained through the following formula:

VO2 max = Distance in meters x 0.0136 + 45.3

3.2.2. Counter Movement Jump Test

Subjects' explosive power was assessed using the countermovement jump test (CMJT). The subjects first stand on the screen and, immediately after the initial downward movement, jump straight up while the hands are on the hip joint during the jump. The test was performed three times with a minute of rest between each jump, and the best record was calculated. The height of the center of gravity from the ground was calculated by the following formula (14):

$$Jump \ height \ = 9.81 \times \frac{Flight \ time^2}{9}$$

3.3. Supplementation

Participants were assigned to their supplements in a randomized, double-blinded manner. The subjects ingested either five g/d beta-alanine (n = 8) or a maltodextrin placebo (n = 8). The total daily dose was split into five individual 1 g doses to be taken along with main meals (breakfast, lunch, afternoon snack, and dinner) and before training. On rest days, another 1 g was to be taken along with brunch. In order to reduce the effect of diet on the study's results, all subjects followed the same diet plan for the last three days of supplementation until after blood sampling (15).

3.4. Blood Samples and Analyses

After ending supplementation and venous blood samples 24 hours before the competition and 48 hours after the match, 3cc of blood was taken from the brachiocephalic artery (Figure 1). Blood samples were centrifuged at 3000 rpm for five minutes using an isolated serum to measure research variables. All of the kits had been bought from Bionik company.

3.4.1. CK

NAC. Kinetic UV. The liquid in serum or plasma was used to measure the creatine kinase. The diameter of the cuvette was 1 cm. The wavelength at 25, 30, and 37°C was set at 340 nm. The sensitivity of this method was 10 U/L.

3.4.2. LDH

Pyruvate. Kinetics UV. DGKC. The liquid in serum or plasma was used to measure the lactate dehydrogenase. The diameter of the cuvette was 1 cm. The wavelength at 25, 30, and 37°C was set at 340 nm. The sensitivity of this method was 1 U/L.



Figure 1. Overview of study design. PG, placebo group; BG, beta-alanine group; CK, creatine kinase; LDH, lactate dehydrogenase; CMJT, countermovement jump test



3.4.3. UREA

Urease-GLDH. Liquid in serum or plasma was used to measure the urea. The diameter of the cuvette was 1 cm. The wavelength at 15, 25, and 37 C was set at 340 nm. The sensitivity of this method was 1 mg/dL.

3.5. Statistical Analysis

The normality of the distribution of the variables was tested using the Kolmogorov-Smirnov test. An analysis of covariance (ANCOVA) was used to assess differences between the groups. Pre-test and post-test changes of each variable were analyzed using a paired-sample *t*-test. Statistical analysis was performed using IBM SPSS statistics (version 22, IBM). P-values < 0.05 were considered significant.

4. Results

Body composition indicators (height, weight, body mass index) were also evaluated (Table 1). Changes in blood indices related to recovery after 21 days of supplementation are reported in Table 2. According to the results, no significant changes were observed in serum creatine kinase, lactate dehydrogenase, and urea levels in the betaalanine group compared to the placebo (P < 0.05). However, in the within-group comparison, significant changes were only observed in the increase of creatine kinase in the beta-alanine group (P < 0.05).

In terms of performance, no significant changes were observed in the maximum oxygen consumption and explosive power in the comparison of the two groups (P < 0.05) (Table 3). However, in the within-group comparison, significant changes were observed in the improvement of the maximum oxygen consumption in both groups, which was higher in the beta-alanine group (P \leq 0.01).

5. Discussion

In this study, we found that beta-alanine had no effect on the reduction of metabolic factors related to recovery, nor did it improve the performance of soccer players.

Previous studies that investigated the effect of betaalanine supplementation on metabolic parameters related to recovery; showed similar findings; Roveratti et al. did not observe a significant impact on reducing creatine kinase index after using beta-alanine supplements (16). Also, Gholami et al. showed that four weeks of BA supplementation had no significant effect on CK in female basketball players (17). In resistance training, the increase in creatine kinase levels may be more effective due to the further destruction of the cell membrane (18).

The mechanism and pattern of changes (increase) of serum total creatine kinase enzyme following aerobic training are mainly due to leakage due to energy loss and instability or damage due to peroxidation of cell membrane phospholipids (19). In the present study, due to the fact that blood sampling was done 48 hours after intense activity, and in the baseline state, the subjects did not perform intense activity 48 hours before the blood sampling, we expected an increase in blood indices, including creatine kinase, compared to the baseline state. Still, because the groups were in the same conditions, we expected that the beta-alanine group would increase less than the placebo, but no significant difference was observed between the groups. The interesting point here was that in the intra-group comparison, we expected the amount of creatine kinase to increase less in the beta-alanine group than in the placebo. Still, we observed that not only did it not increase less, but we also saw a greater increase compared to the control group, which was a significant change.

Our findings did not observe a significant change in serum lactate dehydrogenase concentration. Similarly, Saunders et al. did not report a decrease in plasma lactate dehydrogenase concentration after 24 weeks of beta-

Table 1. Characteristics of Subjects ^a							
Variables	No.	Age, y	Weight, kg	Height, cm	BMI, kg/m ²		
Beta-alanine	8	20.4 ± 0.96	70.83 ± 11.83	178.77 ± 6.5	22.15 ± 1.98		
Placebo	8	19.6 ± 1.42	64.51± 6.16	173.99 ± 4.58	21.37 ± 2.26		

^a Values are expressed as mean ± SD.

Table 2. Blood Variables (Mean ± SD) Before (Pre) and After (Post) the Interventions

Variables	Groups	Pre-test	Post-test	$\Delta^{\mathbf{a}}$	P-Values ^b	P-Values ^c
CK (mg/dL)	Placebo	247.25 ± 138.858	277.50 ± 55.831	30.25	0.093	- 0.061
	eta-alanine	174.63 ± 53.012	248.75± 77.590	74.12	0.016	
LDH (mg/dL)	Placebo	361.75 ± 64.102	370.88±32.458	9.13	0.627	0.782
	eta-alanine	331.25 ± 50.051	345.63±40.977	14.38	0.362	
UREA (mg/dL)	Placebo	29.550 ± 3.46	31.925 ± 3.36	2.375	0.082	0.155
	β -alanine	33.938±6.72	33.675±6.61	-0.263	0.893	

Abbreviations: CK, creatine kinase; LDH, lactate dehydrogenase.

^a Post-test value - pre-test value.

^b Paired *t*-test (within-group differences).

^c ANCOVA test (between-group differences).

Table 3. Functional Variables (Mean \pm SD) Before (Pre) and After (Post) the Interventions

Variables	Groups	Pre-test	Post-test	$\Delta^{\mathbf{a}}$	P-Values ^b	P-Values ^c
Aerobic Power (mL/kg/min)	Placebo	75.56 ± 4.99	64.2 ± 6.67	-11.36	0.020	- 0.155
	eta-alanine	72.43 ± 5.51	64.23 ± 4.66	-8.2	0.001	
Explosive power (cm)	Placebo	0.326 ± 0.66	0.337±0.73	0.011	0.176	0.569
	eta-alanine	0.335 ± 0.67	0.335 ± 0.44	0.00	0.996	

Abbreviations: CK, creatine kinase; LDH, lactate dehydrogenase.

^a Post-test value- pre-test value.

^b Paired *t*-test (within-group differences).

^c ANCOVA test (between-group differences).

alanine supplementation (20). Still, Karimzadehfard et al. showed a significant increase in LDH levels after three weeks of β -alanine and creatine supplementation in response to an exhausting swimming session among elite swimmers (21). But the time of blood sampling in this study was immediately after intense activity. It probably caused a significant difference in the LDH level. Probably the difference in the amount of LDH in this study with our study might be due to the time of blood sampling.

It is likely that the short time interval for blood sampling to measure the lactate dehydrogenase index in the present study is a reason for the ineffectiveness of betaalanine supplementation because the lactate dehydrogenase index was assessed at least 48 hours after a highintensity activity at its maximum level. However, the lactate dehydrogenase index has been observed to peak 72 hours after strenuous exercise (22). On the other hand, increasing muscle phosphocreatine concentration and

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increasing anaerobic enzymes phosphofructokinase, aldolase, and lactate dehydrogenase can be attributed to intense intermittent soccer training. There is a need for more research in this area (23).

Among the other metabolic indicators related to recovery is urea. Still, few studies are available regarding the effect of beta-alanine supplementation on the amount of urea in soccer players. McCormack et al. investigated the impact of a beta-alanine-enriched dietary supplement on increasing physical work capacity in the elderly and did not observe a significant difference in the amount of urea among the groups (24). Also, recently, Durkalec-Michalski et al. After beta-alanine supplementation in highly trained sprinters and endurance athletes, they did not observe a significant decrease in urea (25). Despite the differences in the current research protocol with previous studies, the obtained results were consistent with each other. The lack of control over the players' diet, hot weather, and lack of drinking enough water during the activity may influence the non-significance of this factor.

The results showed that beta-alanine supplementation does not improve metabolic parameters related to recovery, so our hypothesis of faster recovery for the next session is not accepted.

In line with the effect of beta-alanine supplementation on performance, beta-alanine may improve performance in athletes by increasing the amount of carnosine (26). Bachal and Shori showed that beta-alanine improved aerobic indices in weight lifters (27). An increase in maximal oxygen consumption is generally due to an improvement in oxygen transport to skeletal muscle by increasing stroke volume (28), capillary and mitochondrial density (29), and thus increasing oxygen uptake by active forces. Also, the rate of oxidative phosphorylation follows the designed exercises, which such adaptation may be achieved as a result of increasing the number and activity of muscle oxidation enzymes (30). On the other hand, studies have shown that the buffering capacity of hydrogen ions is improved by increasing carnosine due to beta-alanine consumption, which may also be effective in increasing the aerobic capacity of players following beta-alanine consumption (31). Nevertheless, Gharaat et al and Hadi et al. did not observe any improvement in maximal oxygen consumption following acute consumption of beta-alanine supplements (32, 33). In the current research, the nature of the measurement and its time were different from the previous study. Willy-nilly, after a soccer match, people's record in the aerobic capacity test will decrease due to the fatigue caused by the game compared to the primary state where there was no pressure on them. Still, in similar conditions, beta-alanine supplementation can show its effect on the lower reduction of the recorded record. The current research showed that beta-alanine supplementation did not affect improving people's aerobic capacity. However, in the intergroup comparison, there were significant changes in both groups. Because both groups had their high-pressure soccer training during the week, it seems logical that with the adaptations that have occurred, we will see significant changes in both groups. Still, in the supplement group, beta-alanine caused a smaller drop than in the placebo group. Here is what the effect of this supplement is showing.

The findings of the present study show that betaalanine supplementation had no significant effect on improving the explosive power of players. Consistent with this study, Carpentier et al., following the effect of beta-alanine supplementation and plyometric exercises on women and men athletes, achieved consistent results with the present study. They did not observe a significant difference in explosive power between the groups after two months of training (twice a week) and supplementation (34). Still, Rosas et al. also examined the effect of plyometric training and beta-alanine supplementation on maximal-intensity exercise and endurance in female soccer players. They achieved inconsistent results with the present study, such that the beta-alanine group experienced an improvement in explosive power compared to the control group (35). Plyometric training to improve the explosive power in this study can be one of the reasons for inconsistency with the present study. In general, the probable reasons for not being significant in this index in the current research are rooted in the fatigue of alphamotor neurons, holding the test immediately after playing 90 minutes of football, as well as high blood H+ and its effect on calcium release.

The present study has some limitations, such as not performing muscle biopsies to assess muscle carnosine levels accurately. Participants did not follow the same standard diet. However, each player was asked to follow a similar diet daily and have assessments before performing each step. A final limitation of our study would be the small sample size, raising the possibility of a type II error. Therefore, considering the above, future research can effectively provide more accurate reports on the positive effect of beta-alanine on athletes.

5.1. Conclusions

The results of this study showed that beta-alanine supplementation had no effect on the amount of blood indicators related to recovery, but it may improve the aerobic performance of players. Considering the important role of maintaining aerobic capacity in football, especially in high-pressure matches with consecutive games, coaches can be advised to use beta-alanine supplements to improve athletes' performance.

Footnotes

Authors' Contribution: Study concept and design: Aeen Moniri, Alireza Safarzade; and Amir Esamaeeli; Analysis and interpretation of data: Aeen Moniri, and Alireza Safarzade; Drafting of the manuscript: Aeen Moniri; Critical revision of the manuscript for important intellectual content: Aeen Moniri, Alireza Safarzade, and Amir Esamaeeli; Statistical analysis: Aeen Moniri Alireza Safarzade, and Amir Esamaeeli.

Conflict of Interests: There is no conflict of interest.

Ethical Approval: IR.UMZ.REC.1397.078.

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Informed Consent: Written informed consent was obtained from each participant after the study details were shared in depth with them.

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