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Original Research

Eight Weeks of Resistance Training Supplemented with Beta-Alanine and Sodium Bicarbonate Increased Muscle Cross-Sectional Area

Jamil Jafari Pouresmaeili*[1], Saied Jalal Aboodarda†[2], Hamid Rajabi‡[1]

[1]Department of Exercise Physiology, Kharazmi University, Iran; [2]Faculty of Kinesiology, University of Calgary, Calgary, Canada

*Denotes student investigator, †Denotes early-career investigator ‡Denotes established investigator

Abstract

International Journal of Exercise Science 19(2): 2005, 2026. Sodium bicarbonate (SB) and beta-alanine (BA) can enhance high-intensity exercise performance. This study examined the effects of eight weeks of resistance training combined with these supplements on muscle cross-sectional area (CSA), serum myostatin and interleukin-15 concentration, and upper- and lower-body muscular strength and power. Twenty active males (age: 18-35) were divided into supplement (SUP) and placebo groups (PL; each group n=10). Participants undertook eight weeks of supervised resistance training, including 3 sessions × 3 sets × maximum repetitions at 70% of 1RM. Supplementations included 3- 6 g/day BA and 4-6 g/day SB administered for 8 weeks. Exercise volume, calculated as the total number of repetitions completed in each session, increased to a greater extent following SUP (~29%) than PL (~13%) groups (P=0.014). Central segment of quadriceps CSA, measured using magnetic resonance imaging, demonstrated a greater increase following SUP compared to PL for both right ((~21%; pre to post: 452.47±55.98 to 548.21±73.92 mm^3) vs. PL (~5%; 443.81±28.74 to 466.42±31.91 mm^3)) and left ((~24%, 456.81±59.77 to 566.71±82.62 mm^2) vs. PL (~9%; 438.12±41.54 and 479.06±49.91 mm^3)) legs (all P<0.01). However, interleukin-15 concentrations decreased following both conditions (time effect: P<0.001). Also, no difference was observed between groups for the increased 1-RM squat, bench press, and countermovement jump (time effects: P<0.05). The results suggest that co-ingestion of BA and SB may enhance the central segment of the CSA in the quadriceps muscle, possibly by increasing exercise volume. This could be a potential approach in promoting muscle hypertrophy in athletic and clinical rehabilitation settings.

Keywords: Weight training, muscle hypertrophy, strength, exercise volume, supplementation

Introduction

Supplementation aimed at enhancing exercise performance is a widely applied practice in athletic settings.[1] Among different supplements, sodium bicarbonate (SB) and beta-alanine (BA) have been shown to enhance high-intensity exercise performance.[2,3] The primary mechanism of action for these supplements involves buffering intracellular hydrogen ions (H^+) produced by the glycolytic pathway activity during intensive exercise.[4] While BA - a non-essential, non-proteinogenic amino acid - provides its buffering capacity by increasing muscle carnosine, SB acts by transporting lactate and H^+ through the blood, leading these metabolites outside of the exercising muscles.[5] Buffering of H^+ mitigates exercise-induced neuromuscular function impairment and consequently postpones muscle fatigue.[6] Previous investigations have applied

*Corresponding Author: Hamid Rajabi; Email: hrajabi@khu.ac.ir

co-supplementation of BA and SB and shown its positive impact on the exercise performance of tasks lasting 30 s and 10 min.[7,8] However, the effect of these supplements on the development of muscle cross-sectional area (CSA) and maximal muscle strength and power following a resistance training program has remained under-investigated. While BA and SB do not directly participate in the protein synthesis signaling cascade, the combined intracellular and extracellular buffering capacity of BA and SB may uniquely enhance resistance training volume, potentially amplifying muscle CSA and performance gains.[9]

Indeed, previous investigations have explored the effects of BA and SB on cycling,[7] exercise volume,[7] and performance. However, limited and contradictory studies have investigated the effects of these supplements on resistance exercise outcomes. For instance, Hoffman et al,[10] and Maté-Muñoz et al,[11] investigated the impact of 10 and five weeks of BA supplementation, respectively, and demonstrated the beneficial effects of supplementation on training volume, as well as maximal strength and power development. Carr et al,[12] also found an increase in total repetitions during resistance exercise with SB supplementation. Notably, these studies did not assess changes in muscle cross-sectional area (CSA), a critical measure of muscle hypertrophy. de Camargo et a.,[13] addressed this gap by examining the effect of BA supplementation (6.4 g) on muscle thickness over eight weeks of resistance training. However, these investigators did not find an effect of supplementation on muscle thickness (measured via ultrasound), potentially due to unchanged exercise volume. In this context, a plausible question is whether co-injecting BA and SB could enhance exercise volume and consequently improve muscle CSA. Using an accurate method of assessing muscle CSA, such as magnetic resonance imaging (MRI), can delineate the effect of these supplements on muscle hypertrophy, a key resistance training adaptation in athletic and clinical settings.

Given that muscle CSA serves as the primary macroscopic outcome of hypertrophy in the proposed supplementation protocol, elucidating the underlying molecular mediators□such as the anabolic myokine interleukin-15 (IL-15) and the catabolic regulator myostatin□provides mechanistic insight into how enhanced exercise volume from BA and SB co-ingestion may drive greater hypertrophic adaptations.[14] IL-15 is recognized as an anabolic biomarker that promotes muscle hypertrophy in muscle cells.[15] Riechman et al, [16] demonstrated that IL-15 increased significantly in response to acute resistance training in young men and women. Conversely, myostatin is one of the key negative regulators of skeletal muscle growth, where inactivation of this myokine during resistance training can lead to increased muscle hypertrophy.[17] Prior evidence has demonstrated an association between myostatin reduction and increased muscle CSA.[18] Despite these findings, limited empirical evidence exists on the potential alterations in IL-15 and myostatin in response to resistance training combined with BA and SB supplementation. Therefore, further studies are required to investigate whether co-ingestion of BA and SB may enhance resistance exercise volume and alter IL-15 and myostatin concentrations, the parameters that can contribute to the development of muscle CSA.

Therefore, the present study aims to examine the effects of eight weeks of resistance training combined with BA and SB supplementation on the hypertrophic changes (i.e., muscle CSA) of quadriceps and pectoral muscles, as well as serum myostatin and interleukin-15 levels, upper and lower body muscular strength and power in healthy, recreationally active men. We hypothesize that resistance training with BA and SB supplementation can increase exercise volume and consequently enhance muscle CSA, strength, and power.

Methods

Participants

Based on an a priori sample size calculation (= 0.05, Power = 0.8) performed in G*Power (version 3.1.9.6) derived from a study that investigated the effect of 5-week strength training combined with beta-alanine on different physiological responses, including the number of repetitions completed per set.[11] Twenty healthy young males who had previously engaged in mild to moderate resistance training within the last six months participated in the study (Table 1). None of the participants had a history of neurological, cardiovascular, or metabolic diseases, nor did they use any supplements 12 months before this study. Participants refrained from ingesting any additional drugs or dietary supplements and participating in any other resistance training program across the eight weeks of the study. Ethics approval was obtained from the local ethics board (IR.KHU.REC.1403.018) and the study was conducted in accordance with the Declaration of Helsinki (without registration). Also, this research was carried out in accordance with the ethical standards of the *International Journal of Exercise Science.*[19]

Table 1. Description (mean ± standard deviation) of participants' characteristics.

Variable	Placebo) n=10)	Supplement) n=10)
Age (years)	22 ± 2.15	23 ± 1.93
Weight (kg)	75 ± 10.802	77 ± 9.938
Height (cm)	179 ± 4.351	177 ± 6.909
Body Mass Index (kg/m²)	23 ± 2.87	24 ± 3.50

Protocol

Experimental procedure and supplement prescription.

After completing the consent form and training history questionnaire, participants were randomly divided into two groups (each n=10): the placebo group (PL) and the supplement group (SUP). BA supplementation was administered three times a day (morning, noon, and evening) alongside everyday meals (breakfast, lunch, and dinner) for eight weeks (56 days).[20] A progressive increase in supplementation was administered, whereby BA (Doobis Nutrition, IR) was increased from 3 to 6 g/day (one gram increase every two weeks in capsule content) from the first to the last two weeks of the program. For SB (Doobis Nutrition, IR), the dosage increased from 4 grams during the first four weeks and 6 grams during the second four weeks. The SB was ingested 30 minutes before each training session.[21] To minimize the side effects of consuming BA and SB (e.g., the tingling sensation induced by BA or diarrhea caused by SB), the doses of BA and SB were progressively increased. The PL group received placebo capsules filled with dextrose (Merck KGaA, Germany). For compliance tracking, participants were asked weekly—before each training session—and they confirmed full adherence, reporting no skipped or missed doses. Participants were informed that the study explored the effect of supplements on some physiological factors without further information on whether they were receiving supplements or a placebo. All participants resided in the university dormitory and received standardized meals for lunch, dinner, and breakfast provided by the university. The amount of protein consumed each week was measured using a nutritional value table (converting raw food to cooked). The protein intake for each participant ranged from 305 to 315 grams per week (approximately 0.8 grams per kilogram of body weight per day). Regarding side effects, some participants experienced diarrhea and paresthesia in the supplement group. Diarrhea resolved after week 2, and paresthesia diminished after week 4. Participants who were in the placebo group did not have any side effects.

Exercise prescription.

After completing a 1-repetition maximum (1-RM) test for squat, leg extension, bench press, and shoulder press, to assess upper-body endurance, participants performed a maximum pull-up test, including maximal body-weight pull-ups until task failure. Thereafter, both groups participated in an eight-week supervised resistance training program. To familiarize participants with the program, a lower intensity (40% 1-RM) of the exercises was applied in the first week. In the subsequent weeks, both groups performed three sets of squats, leg extensions, bench presses, and shoulder presses at 70% of 1RM as well as maximum pull-ups to task failure with 60 s of rest between each set and 120 seconds between exercises.[22] Task failure was defined as the inability to perform a full repetition without spotting. The sequence of exercises included squats, bench presses, leg extensions, shoulder presses, and pull-ups for all participants. This order was selected to increase rest time between the two lower (squat and knee extension) and the two upper body exercises (bench press and shoulder press). All workout sessions were conducted in the evening (5-7 PM) on Saturdays, Mondays, and Wednesdays. Four weeks after the first training session, participants were re-evaluated with a 1RM test to adjust weights for the following four weeks while maintaining the expected intensity of 70% of 1RM. The number of repetitions completed in each set was recorded to determine the training volume.

Measurements.

Exercise volume was calculated as the total number of repetitions completed for six exercises (squats, leg extensions, bench presses, shoulder presses, and pull-ups) in the first session of the second week and the last session of weeks 4, 6 and 8. The first week of training was intentionally skipped because this week served as a familiarization period, during which participants performed exercises at lower intensities (40% of 1-RM). Therefore, to assess the effect of supplementation on exercise volume, the counting of repetitions began in the second week of the program. Since the relative exercise intensity for the two groups within the second and the last week of the program was similar (70% of each individual's 1-RM), the number of repetitions could reflect changes in exercise volume.

MRI scans.

The CSA was assessed using a 3-Tesla MRI machine (Siemens Healthcare, Erlangen, Germany). The participant rested in a supine position for around 10 minutes prior to any scan for both the pectoralis and quadriceps muscles. In both MRI sequences, cross-sectional images in the axial plane were obtained 1 day before and 1 day after the training program. Two different sequences were run in each testing session. In the T1-weighted used for imaging, the acquisition parameters were TE 110 ms, TR 5723 ms, number of signal averages: 3, FOV 48.5 (x-direction) × 39.4 (y-direction cm, voxel size 0.95 × 0.95 × 10 mm, with a scan time of about 5 min.

Image analysis was conducted using the latest version of the 3D Slicer software (3D Slicer is an open-source software platform developed by the Surgical Planning Lab at Harvard Medical School, United States). The use of this software for assessing muscle cross-sectional area has been validated in a previous study.[23] To measure muscle volume, with values recorded in square centimeters, the quadriceps muscle was divided into approximately ninety slices, and the proximal, central, distal, and total muscle volumes were assessed for the left and right quadriceps muscles, between the greater trochanter and 2 centimeters above the patella, by averaging forty slices. The pectoralis muscle on the left and right sides was divided into approximately eighty slices, and muscle volume was assessed by averaging thirty slices in the belly of the muscle (Figure 1).

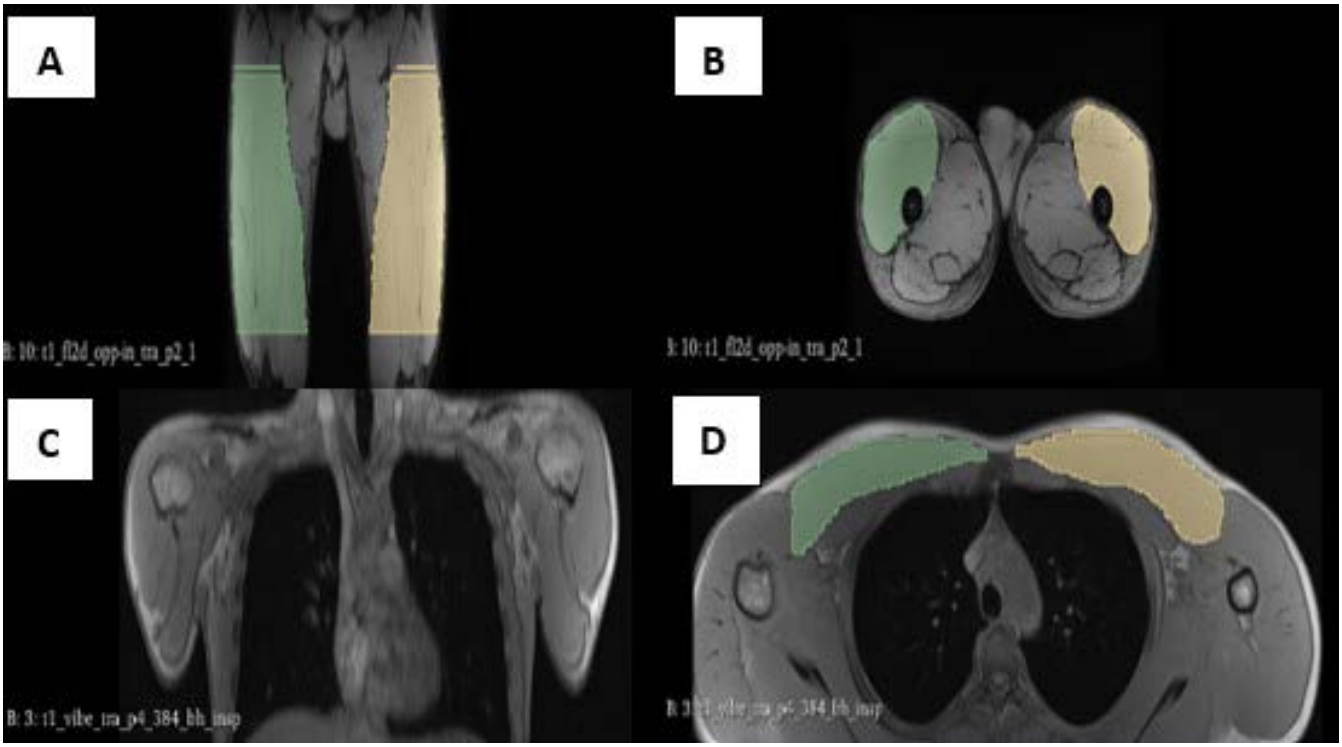

Figure 1. The MRI analysis of the quadriceps (panels A and B) and pectoral (panels C and D) muscles. The proximal, central, distal, and total muscle volume was assessed for the quadriceps muscles (between the greater trochanter of the femur and 2 cm above the patella) and the total pectoralis muscle on the left and right sides of the body. Panels A and C depict axial T1-weighted images of the thigh and pectoralis muscles, and panels B and D explain coronal images from the thigh and pectoralis muscles, respectively.

Blood Biomarkers.

Twenty-four hours before the beginning of the resistance training program and after the final training session, 2 milliliters of blood were drawn from the median cubital vein in a non-fasted state for the assessment of Interleukin-15 and myostatin levels. The measurements were conducted in the laboratory using the Zell Bio kit (Germany) and the ELISA method.

The lower and upper body strength were assessed by measuring 1-repetition maximum (1-RM) squat and bench press tests, respectively, using a Smith machine (Akoosport, IR). Each participant completed as many trials as required to achieve 1-RM, with a 2-minute rest between attempts.

Muscular power was tested using a countermovement jump. Standing against a wall with totally extended and stretched arms, the participant's height was measured. Then, a vertical jump test (Sargent jump) was performed for one repetition at maximum effort, marking the distance between two points. The jump height was calculated based on the difference between these points in centimeters. Each participant completed two trials with a 2-minute rest between attempts, and the best score was recorded. This test's use in evaluating lower-body muscular power has been validated in a previous study.[24]

The participants' dominant leg was assessed using the Leg Dominant Determination questionnaire.[25] Additionally, the "Edinburgh Handedness" questionnaire was used to evaluate the dominant hand of individuals.[26]

Statistical Analysis

The Shapiro-Wilk test was used to evaluate the normality of the data. Repeated measure analysis of variance (ANOVA) with independent samples and Bonferroni post hoc test were utilized to test exercise volume across four time points (i.e., weeks 2, 4, 6, and 8) and two groups (SUP and PL). For all other measurements, including quadriceps and pectoralis muscle CSA, serum myostatin and interleukin-15 concentrations, muscle strength and power metrics, separate repeated measure ANOVAs with independent samples and Bonferroni post hoc test were performed for two time points (pre- vs. post-training) and two (SUP and PL). Effect sizes (η^2 for ANOVA and Cohen's d for pairwise comparisons, classified as small [0.2], medium [0.5], or large [0.8]) were calculated. SPSS version 26.0 (IBM Corp., Armonk, NY, USA) was used to analyze the data, and significance was set at $P < 0.05$.

Results

For exercise volume (number of repetitions completed), significant time ($F_{3,54} = 49.06$, $P < 0.001$, $\eta^2 = 0.732$) and interactions ($F_{3,54} = 5.80$, $P = 0.002$, $\eta^2 = 0.244$(were observed with 29% and 13% increase in exercise volume following across eight weeks for SUP and PL programs, respectively (Figure 2). The exercise volume was greater in the last week of SUP vs. PL condition ($P = 0.005$, $d = 0.993$).

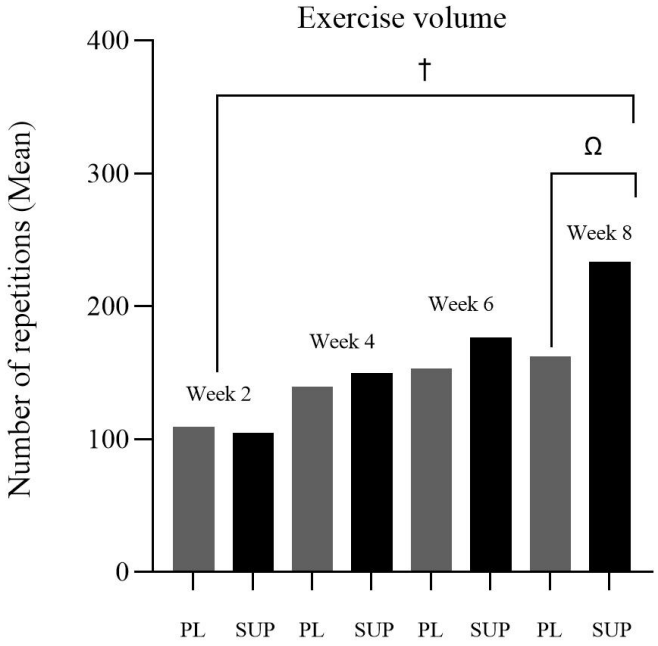

Figure 2. The exercise volume between the two groups. SUP: Supplementation group; PL: Placebo group. Ω Different than PL at week 8; † Significant time effect.

Muscle Cross-Sectional Area

The total cross-sectional area of the left quadriceps demonstrated no time ($P = 0.089$) or interaction effects ($p = 0.279$). However, there was a time effect for the total CSA of the right quadriceps ($F_{1,18} = 71.07$, $P < 0.001$) without any interaction effect ($P = 0.616$), where this measure increased by 14.3% and 14.1% following the SUP and PL programs, respectively (Figure 3).

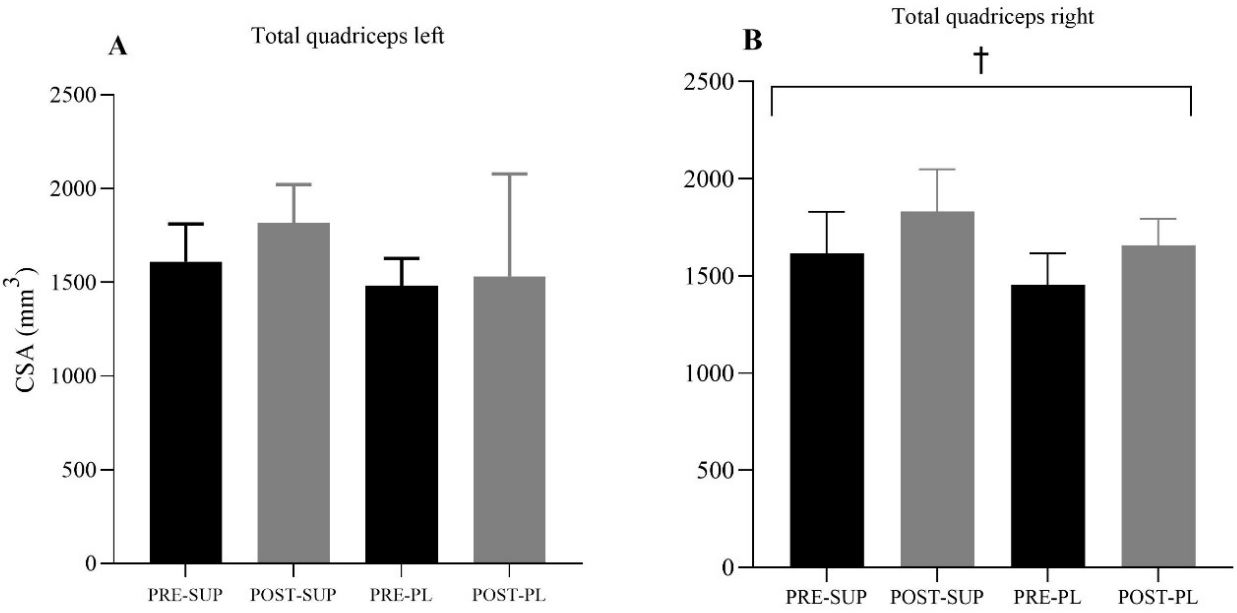

Figure 3. The changes in total CSA of right and left quadriceps from pre- to post-training between two groups. SUP: Supplementation group; PL: Placebo group. † Significant time effect.

The proximal CSA of the left quadriceps showed a time effect ($F_{1,18}$ = 25.46, P < 0.001) without an interaction effect (P = 0.279), where this parameter showed a 12% and 9% increase for the SUP and PL, respectively. Similarly, the proximal CSA of the right quadriceps illustrated a time effect ($F_{1,18}$ = 44.49, P < 0.001) without an interaction effect (P = 0.760). There was a 12% and 11% increase for the SUP and PL, respectively.

The central CSA of the left quadriceps demonstrated time ($F_{1,18}$ = 25.35, P < 0.005, η^2p = 0.585) and interaction effects ($F_{1,18}$ = 5.299, P < 0.034, η^2p = 0.227) with 24% and 9% increases for the SUP and PL groups, respectively (P = 0.883, d = 0.004). Similarly, the central CSA of the right quadriceps showed time ($F_{1,18}$ = 27.47, P < 0.001, η^2p = 0.604) and interaction effects ($F_{1,18}$ = 10.48, P < 0.001, η^2p = 0.368) with 21% and 5% increases following SUP and PL programs, respectively (P = 0.990, d = 0.005).

The distal CSA of the left quadriceps showed a time ($F_{1,18}$ = 6.68, P < 0.019) without an interaction effect (P = 0.184) with 9% and 3% increases following SUP and the PL programs, respectively. The distal CSA of the right quadriceps demonstrated a time effect ($F_{1,18}$ = 4.45, P < 0.049) without an interaction effect (P = 0.338), with 8% and 3% increases following SUP and PL groups, respectively (Figure 4).

The total CSA of the left pectoral muscle showed a time effect ($F_{1,18}$ = 218.32, P < 0.005) without an interaction effect (P = 0.331). This measure demonstrated 68% and 78% increases following SUP and PL programs, respectively. The total CSA of the right pectoral muscle demonstrated a significant time effect ($F_{1,18}$ = 154.64, P < 0.001) without an interaction effect (P = 0.604) where this measure showed 59% and 67% increases following the SUP and PL groups, respectively (Figure 5).

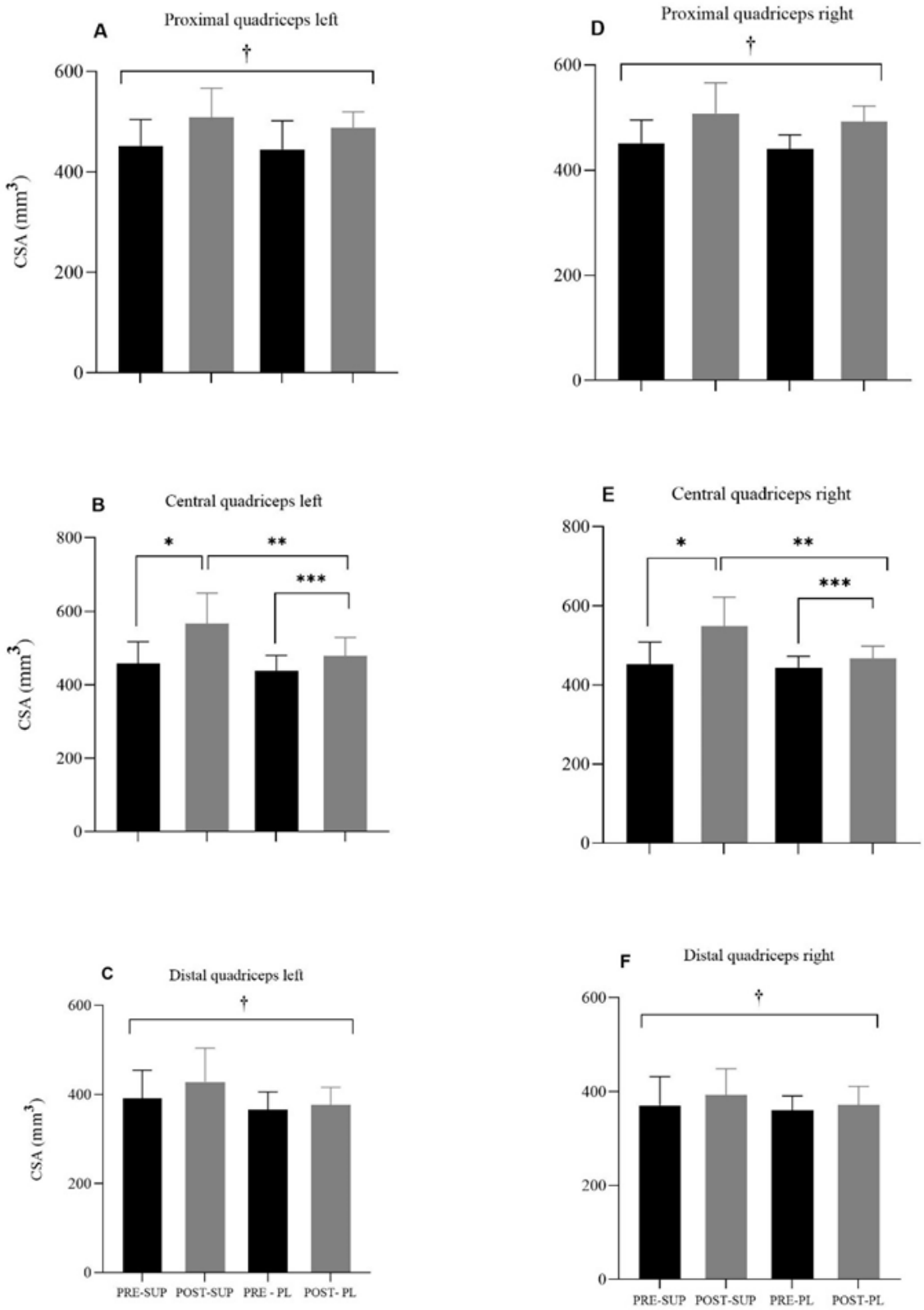

Figure 4. Changes in proximal, central, and distal CSA of the left and right quadriceps muscles from pre- to post-training between the two groups. SUP: Supplementation group; PL: Placebo group. * Different from PRE-SUP; ** Different between SUP and PL post-training; *** Different from PRE-PL; † Significant time effect.

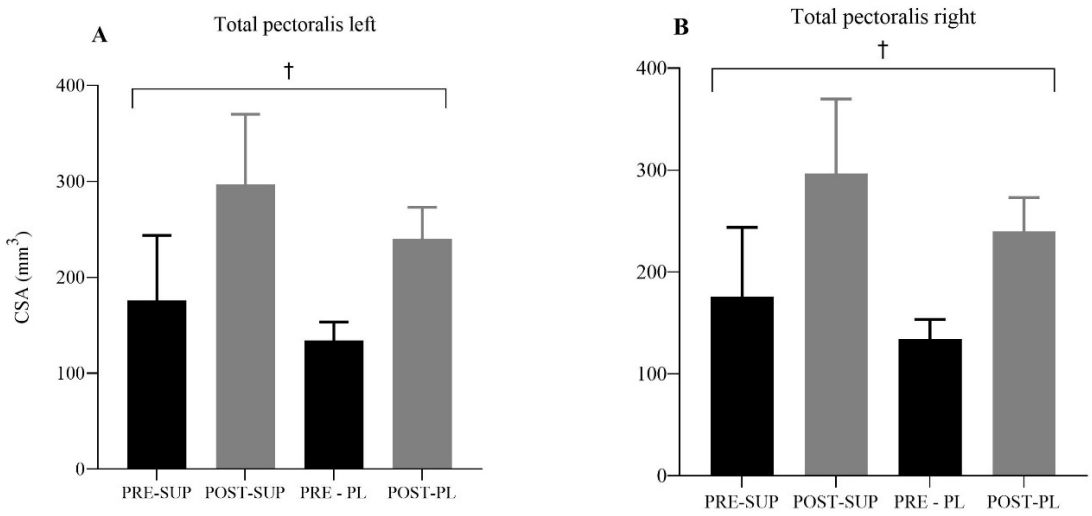

Figure 5. The changes in total CSA of the right and left pectoralis muscles from pre- to post-training between the two groups. SUP: Supplementation group; PL: Placebo group. † Significant time effect.

Serum IL-15 and myostatin

The serum IL-15 showed a time effect ($F_{1,18}$ =7.65, P = 0.013) without an interaction effect (P > 0.448), where there was a 19% and 31% decrease in this measure following SUP and PL groups, respectively. For myostatin, a significant time effect ($F_{1,18}$ =198.42, P < 0.001) was observed without an interaction effect (P = 0.885). There were 28% and 30% decreases in myostatin concentration for the SUP and PL groups, respectively (Table 2 and 3).

Table 2. Pre and post 8 weeks and blood measurements for the SUP and PL groups (mean ± standard deviation).

Variables	Pre	Post 8 weeks	MD [95 % CI]	time *p-value*	time*group *p* value	group
			myostatin (ng/ml)			
SUP	149.31 ± 5.49	106.45 ± 8.57	42.86 [36.35 to 49.36]	0.005	0.88	0.269
PL	143.67 ± 22.86	99.910 ± 11.52	43.76 [31.46 to 49.36]			
			Interleukin-15 (ng/ml)			
SUP	133.50 ± 53.20	107.29 ± 6.38	26.21 [-12.34 to 67.76]			
PL	147.94 ± 68.60	101.290 ± 8.27	46.65 [1.25 to 92.04]	0.013	0.448	0.774

Note: SUP: Supplementation group; PL: Placebo group; CI: Confidence interval.

Table 3. Pre and post 8 weeks functional measures for the SUP and PL groups (mean ± standard deviation).

Variables	Pre	Post 8 weeks	MD [95 % CI]	time *p* value	time*group *p* value	group
			Lower muscular strength (kg)			
SUP	48.50 ± 14.69	67.0 0 ± 15.59	- 0.100 [- 11.71 to 11.51]	0.005	0.460	0.460
PL	34.20 ± 7.71	54.80 ± 11.26	-20.60 [- 25.55 to - 15.64]			
			Upper body Muscular strength (kg)			

SUP	72.80 ± 11.39	115.50 ± 20.56	- 42.70 [- 53.37 to - 32.03]	0.005	0.94	0.005
PL	43.50 ± 17.41	85.60 ± 18.28	- 42.10 [- 57.13 to -20.07]			
			Muscular power			
SUP	18619.50 ± 761.19	21797.00 ± 1079.82	-3177.5 [-3523.91 to -2831.08]	0.005	0.907	0.037
PL	18279.00 ± 523.94	21423.30 ± 662.22	-3144.3 [- 3678.85 to -2609.74]			

Note: SUP: Supplementation group; PL: Placebo group; CI: Confidence interval.

Muscle strength and power

After eight weeks of resistance training, the lower body muscular strength measured via 1-RM squat test demonstrated a time effect ($F_{1,18}$=197.43, P < 0.005), without an interaction effect (P = 0.460), where 7% and 2% increases were observed following SUP and PL programs, respectively. The upper body muscular strength measured via the bench press test demonstrated a significant time effect ($F_{1,18}$ =108.317, P < 0.005), without interaction effect (P = 0.942), with 22% and 21% increases following SUP and PL groups, respectively. Similarly, lower body muscular power showed a significant time effect ($F_{1,18}$ = 504.041, P < 0.005), without an interaction effect (P = 0.907). There was a 3% and 2% increase for SUP and PL groups, respectively (Table 3).

Discussion

The primary objective of this study was to investigate the effects of BA and SB combined with resistance training on exercise volume and CSA. The results indicate that the supplementation administered over the eight weeks significantly enhanced exercise volume and the central segment of both the right and left quadriceps CSA. However, this intervention did not yield significant changes in the total quadriceps or pectoralis CSA, nor did it result in any significant difference between the two conditions for changes in myostatin and IL-15 concentrations, upper and lower body maximal strength, and the lower body power. These findings indicate that the co-ingestion of BA and SB may enhance resistance training volume, potentially amplifying IL-15 release and reducing myostatin, thereby promoting greater muscle CSA through synergistic effects on myokine-mediated hypertrophy pathways. This study investigates these alterations to elucidate their role in BA and SB-driven muscle adaptations.

The effect of BA and SB co-ingestion on exercise volume

Combined ingestion of BA and SB enhanced exercise volume. This effect is likely attributed to improved muscle and blood buffering capacity, facilitating the removal of intracellular protons (H+) during intense muscle contractions.[27] This observation corroborates the findings of prior investigations that demonstrated the positive effect of BA and/or SB ingestion on resistance exercise volume.[10,28] Contrary to these studies, however, some investigators did not observe an increase in resistance exercise volume, which could be partly due to the shorter duration (e.g., 4 weeks) of the resistance training program,[29] selection of resistance-trained participants,[13] or sole ingestion of BA or SB. Nevertheless, findings in the current study suggest that a longer-term use of combined BA and SB could improve exercise volume in recreationally active participants.

The effect of BA and SB co-ingestion on the muscle cross-sectional area

The improved training volume with combined BA and SB was concomitant with a significant increase in the central segment of the right and left quadriceps CSA compared to the PL

condition. To the best of our knowledge, this is the first study that has used MRI to explore nuanced morphological adaptations of different segments of exercised muscles combined with co-ingestion of BA and SB, and demonstrates the positive impact of these supplements on increased central segment of muscle CSA of the lower limb muscles. This increase could be attributed to enhanced time under tension - an important variable in stimulating muscle hypertrophy signaling pathways. This relationship has been proposed as a significant driver of anabolic processes (metabolic stress).[30] Additionally, higher exercise volume can increase the total mechanical tension exerted on fibers to adapt and grow larger. In concert with this explanation, Schoenfeld et al,[31] and Heiki et al,[9] reported significantly greater gains in muscle CSA with higher exercise volumes**.** It is however worth noting that the increased quadriceps CSA was only evident in the central segment of the muscle. No changes were observed in the total, proximal, or distal segments of the quadriceps CSA. In addition, no change was observed in the CSA of the pectoralis muscle. This may be due to the central region of the muscle having undergone a higher degree of training stress.[32] In line with this hypothesis, prior studies have suggested that muscle CSA does not change uniformly across various muscle groups. Indeed, Abe et al,[33] suggested that changes in muscle CSA did not occur uniformly across individual muscles or regions of the body, which may support our findings. Overall, the current study indicates that the co-ingestion of BA and SB supplements may enhance muscle CSA, and this observation is facilitated by segmental analysis of the quadriceps muscle using MRI. This effect is likely mediated through increased training volume, which can lead to greater muscular adaptations.

Alterations in myostatin and IL-15 in response to co-ingestion of BA and SB.

Myostatin decreased in response to the resistance training, without any difference between the two groups. The decline in myostatin was in line with prior studies; however, it is worth clarifying that no study to date has explored the effect of BA and or SB ingestion on this measure. Accordingly, the comparison of our results is limited to prior studies that investigated the effect of resistance training alone. For instance, Bagheri et al,[34] examined the level of myostatin after 8 weeks of resistance training and showed a comparable decrease in myostatin in all training groups. In another study, Walker et al,[35] showed that plasma myostatin level reduced by roughly 20% in participants after 10 weeks of resistance training. The decrease in myostatin has been suggested to play a role in creating an anabolic environment in exercised muscles (increased growth-promoting factors and decreased growth-inhibiting factors) and increased muscle CSA in response to resistance training programs. However, the reduction in myostatin was similar in both groups, indicating that the supplementation and increased exercise volume did not alter the release of this muscle growth inhibitor. However, a caveat with our research method is that blood samples were collected in a non-fasted state, which may have affected circulating cytokine concentrations due to diurnal and nutritional variability. Moreover, IL-15 and myostatin were measured only at baseline and post-intervention. Given that prior literature suggests acute and chronic responses may differ, this approach may not fully capture the dynamic changes in these markers.

In addition to myostatin, IL-15 levels also decreased in response to eight weeks of resistance training. This observation was contradictory to some of the prior studies that showed increased IL-15 in response to acute resistance exercise;[16] however, it is worth noting that there is a difference in the acute versus chronic effect of training on this measure. For instance, Khalaf et al,[36] conducted a systematic review and meta-analysis with 15 studies involving 411 participants and 12 studies involving 899 participants for the acute and chronic effects of resistance training on IL-15 responses, respectively, and demonstrated that acute, but not chronic exercise training,

increased circulating IL-15 concentrations immediately after exercise. The potential mechanism for this observation is unclear, particularly as secretion of IL-15 occurs from several tissues, and the regulation of IL-15 occurs by numerous receptor isoforms.[37] By assessing IL-15 at 24 h, we aimed to detect any persistent modulation of this anabolic myokine that might reflect cumulative hypertrophic signaling over the training period, distinct from the acute post-exercise spike reported in prior literature. Therefore, it is recommended that future research assess the levels of myostatin and IL-15 immediately after resistance training to provide better insights into the mechanisms involved in muscle growth following different resistance training protocols. As mentioned earlier, to the best of our knowledge, this was the first study that explored the effect of combined BA and SB supplementation on serum cytokines. Therefore, future studies should assess the possible effects of BA and SB on these blood factors.

The effect of supplementation on muscular strength and power

Our results showed a time effect for 1RM and muscle power indicating that the co-ingestion of BA and SB did not alter muscle strength and power development differently than PL group. In line with our finding, Outlaw et al,[38] measured the effects of BA supplementation and resistance training on Wingate peak power, and maximal strength at baseline, 4 weeks, and 8 weeks in collegiate women and showed that maximal strength did not significantly improve after 8 weeks. Similarly, Kendrick et al,[39] demonstrated increased muscle carnosine but yielded no greater improvement over placebo in whole body strength after 10 weeks of resistance training coupled with 6.4 BA consumption. For muscle power, Maté-Muñoz et al,[28] demonstrated no effect of BA ingestion during 5-week resistance training on power production capacity. Similarly, Varovic et al,[40] assessed the effects of SB ingestion on muscular strength and power, and reported no additional increase in muscle strength and power in the intervention group. Contrary to these observations, Hoffman et al,[10] showed that resistance training and BA improved lower-body muscular endurance and lower-body power in men after 3 weeks. The reason for the disparity in findings remains unclear, but the lack of strength and power gains in this study might be attributed to the resistance training background that the participants had within the six months before this research. Indeed, prior resistance training could mitigate the potential for rapid neural adaptations, which often drive significant strength gains in novice trainees. Additionally, substantial hypertrophic gains, necessary for further strength improvements, typically require training durations longer than 8 weeks. Thus, participants in this study might have needed a longer training program to achieve significant improvements in muscle strength and power.

Methodological considerations

This study has several limitations that should be acknowledged. First, the relatively small sample size may have limited the statistical power to detect between-group differences. Second, while participants were blinded to both the research question and the supplement or placebo they received, the supplement group experienced paresthesia (a tingling sensation stemming from beta-alanine), which might have compromised blinding for this group. Therefore, participants in this group might have suspected they were receiving the active supplement, potentially influencing their motivation to perform more repetitions. However, participants in both groups remained unaware of the research question and the existence of another group for comparison. Therefore, caution is warranted when interpreting the physiological versus psychological effects of the supplement

on increased exercise volume. In addition, researchers were not blinded to group assignments. This single-blinded design might have introduced potential bias; however, investigators adhered strictly to predefined data collection and data analysis protocols to minimize contamination of the findings. Dietary protein intake was approximately 0.8 g/kg/day, provided by dormitory meals, which may be suboptimal and potentially limit muscle hypertrophy responses. Therefore, future studies should test the combined BA and SB supplementation at higher levels of protein ingestion (e.g., 1.2-1.6 g/kg/day). Finally, with respect to biomarker analyses, blood samples were collected in a non-fasted state, which might have affected circulating cytokine concentrations due to diurnal and nutritional variability. In addition, the delayed measurement of IL-15 may not represent a precise marker of training-induced adaptations. Thus, future studies should consider the immediate measurement of IL-15 to reflect its acute changes to resistance exercise.

Overall, the results of the present study indicate that 8 weeks of resistance coupled with BA and SB supplementation might significantly enhance training volume. This effect may be attributed to the improvement in the muscle and blood buffering capacity, which can delay muscle fatigue. Furthermore, a notable increase in the central segment of quadriceps muscle CSA was observed, which may be attributed to increased training volume and time under tension. This intervention however did not lead to notable changes in the pectoralis muscle CSA, myostatin and IL-15 levels, upper and lower body maximal strength, or lower body power compared to the placebo condition. Further research with a larger sample size is needed to confirm this finding and investigate the underlying mechanisms.

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