



## **Beta-alanine Supplementation for Four Weeks Increases Volume Index and Reduces Perceived Effort of Resistance-trained Men: A Pilot Study.**

JÚLIO BENVENUTTI BUENO DE CAMARGO<sup>†1</sup>, FELIPE ALVES BRIGATTO<sup>†1</sup>, RAFAEL SAKAI ZARONI<sup>†1</sup>, MOISÉS DIEGO GERMANO<sup>†1</sup>, RAPHAEL MACHADO DA CONCEIÇÃO<sup>†1</sup>, ALEXANDRE NUNES MARTINS<sup>‡2</sup>, MARCELO SALDANHA AOKI<sup>‡2</sup>, TIAGO VOLPI BRAZ<sup>†1</sup>, and CHARLES RICARDO LOPES<sup>†1,3</sup>

<sup>1</sup>Human Performance Research Laboratory, Methodist University of Piracicaba, Piracicaba, São Paulo, BRAZIL; <sup>2</sup>School of Arts, Sciences, and Humanities, University of São Paulo, São Paulo, BRAZIL; <sup>3</sup>Adventist Faculty of Hortolândia, Hortolândia, São Paulo, BRAZIL

<sup>†</sup>Denotes graduate student author, <sup>‡</sup>Denotes professional author

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### ABSTRACT

*International Journal of Exercise Science* 14(2): 994-1003, 2021. The aim of the present study was to assess the short-term effects of four weeks of beta-alanine supplementation (BA) (6.4 g/day) on the total volume performed and perceived effort of resistance-trained individuals. Sixteen trained men (age:  $27.3 \pm 5.0$  years, height:  $1.78 \pm 0.1$  cm, total body mass:  $84.3 \pm 8.4$  kg, RT experience:  $5.9 \pm 3.3$  years) were allocated in one of the following groups: BA or Placebo (PLA). In addition, during the same period, participants were submitted to a resistance training program. Volume index (VI) and the rate of perceived exertion (RPE) were collected during the experimental period for both groups. Significant increases from the first to the last intervention week in VI were observed only for BA (+6.5%,  $d = 0.61$ ,  $p = 0.04$ ). In addition, supplementation induced a lower mean RPE (BA:  $8.8 \pm 0.5$  AU vs. PLA:  $9.4 \pm 0.3$  AU,  $p = 0.02$ ). In conclusion, four weeks of BA supplementation were able to increase resistance-training volume without affecting the perceived effort of trained men.

**KEY WORDS:** Strength training, supplement, muscle carnosine, training load

### INTRODUCTION

The buffering capacity during high-intensity exercises is enhanced by increased levels of the dipeptide carnosine, which is synthesized in human skeletal muscle from the amino-acids histidine and beta-alanine (BA) (11, 23). The availability of BA is suggested to be a limiting factor in carnosine synthesis, since skeletal muscle cells have a high L-histidine content (1). In this sense, BA supplementation has been shown to significantly increase muscle carnosine content leading to an ergogenic effect for individuals engaged in high-intensity activities (3, 5). Training volume is described to be one of the major determinants of resistance training (RT)-induced adaptations (4, 21). Although not completely elucidated, higher training volumes seem to result in larger strength and morphological adaptations (2). Practitioners aiming to

potentiate RT outcomes by increasing and/or accumulating training volume during a given period of time may benefit from ergogenic aids that induce improvements in muscle performance.

Although scarce, the available evidence points that RT volume seems to be positively influenced by BA supplementation protocols. Hoffman et al. (6) reported that the addition of BA to creatine supplementation induced significant improvements in training volume (bench press and squat exercises) in collegiate football players. Similarly, 30 days of supplementation were able to increase training volume (total number of repetitions performed during a training session) in strength/power athletes (7). More recently, Mate-Munhoz et al. (12) observed that a five-week supplementation protocol induced a higher workload of resistance-trained individuals.

The effects of BA in the perceived effort during high-intensity activities have been also investigated. Briefly, there seems to be some conflicting evidence regarding BA as an effective nutritional supplement to improve the perceived exertion and biochemical parameters related to muscle fatigue. Invernizzi et al. (9), for example, reported that an acute dose of BA (2 g) was able to reduce the perceived effort during a maximal isometric knee extension test of physically active men. Differently, some longitudinal studies (four-week period) observed no effect of supplementation on subjective rating of exertion during Wingate and maximal number of repetitions tests (18, 24). However, evidence regarding the effects of BA on the perceived effort of previously resistance-trained individuals remains under investigated. Therefore, the aim of the present study was to assess the effects of four weeks of BA supplementation on training volume and rate of perceived exertion (RPE) of recreationally strength-trained subjects. Our initial hypothesis was that participants ingesting BA would present larger increases on training volume and reduced RPE values during the experimental period.

## **METHODS**

### *Participants*

Sixteen healthy males (age:  $27.3 \pm 5.0$  years, height:  $1.78 \pm 0.1$  cm, total body mass:  $84.3 \pm 8.4$  kg, RT experience:  $5.9 \pm 3.3$  years) recruited from a resistance-trained population participated in the study. Volunteers performed RT (minimum frequency of once a week) and all exercises adopted in the training intervention and in the strength tests for at least one year before entering the study. In addition, participants stated that they were not consuming any dietary supplements that could enhance performance for a minimum of six months prior to the start of the study and presented minimal one repetition-maximum (1RM) relative to body mass values of 1.25x and 1x total body mass in back squat and bench press exercises, respectively (2). This study was approved by the University's research ethics committee (protocol 2.094.534) and was conducted in accordance with the Declaration of Helsinki and the ethical standards of the International Journal of Exercise Science (14). All subjects read and signed an informed consent document.

### *Protocol*

The study followed a placebo (PLA)-controlled, double-blind design. For the randomization process, participants were organized into quartile blocks based on their 1RM bench press and back squat scores. Then, each quartile was randomized to one of the experimental groups using computer-generated random numbers. Participants were then subsequently allocated to receive either BA or PLA. The experimental period lasted five weeks. During the first week, participants were submitted to a 1RM test in the bench press and back squat exercises. In addition, a ten-repetition maximum test (10RM) was also performed with aims of familiarization with the exercises adopted during the experimental period. The supplementation and training period was performed during weeks two to five, in which the volume index (VI) and RPE were collected.

Participants received either BA (99.9% pure, non-sustained release formula; CarnoSyn™, NAI, San Marcos, California, USA) or PLA (maldodextrin, Neonutri, Pocos de Caldas, Minas Gerais, Brazil) four times per day (four doses of 1.6 g each [total of 6.4 g/day]), separated by 3-4 hours during the entire training intervention period (four weeks) on both training and non-training days (10). Capsules were similar in appearance, weight (400 mg), and taste. Enough supplement for four weeks was provided in an unlabeled and sealed pot separated by a researcher not involved in data collection. Adherence was determined by counting the amount of capsules remaining at the post-supplementation period. A minimum of 85% of compliance was required (19). Eventual side-effects of supplementation (i.e., paresthesia) were individually monitored during the study. In case a participant forgot to take a dose, an additional dose in another period or on a different day should be ingested to complete the total dose of 179.2 g by the end of the study.

The resistance-training protocol adopted was the same for both groups. During each intervention week, participants performed 4 weekly split routine sessions (A - Mondays and Thursdays, B - Tuesdays and Fridays) (pectoralis major, deltoids, and triceps brachii in routine A, quadriceps, latissimus dorsi and biceps brachii in routine B) (Table 1).

Three sets were performed per exercise with a corresponding load of ten maximum repetitions (RM), with 60 seconds of rest between sets and 120 seconds between exercises (20). Participants were instructed to perform each set to the point of momentary concentric muscular failure. If more than ten repetitions could be performed in the last set of a given exercise, increments in the external load ranging from 5% to 10% were implemented in the next training session.

Training routines were supervised by the research team in order to monitor the proper performance and ensure the safety of each participant. Before the training intervention period, all participants underwent two familiarization sessions in order to determine the individual initial training loads for each exercise.

**Table 1.** Training routines performed by both beta-alanine (BA) and placebo (PLA) groups.

Routine A (Mondays and Thursdays)	Routine B (Tuesdays and Fridays)
Barbell bench press	Barbell back squat
Inclined barbell bench press	Leg press
Dumbbell fly	Knee extension
Dumbbell shoulder press	Dumbbell row
Dumbbell shoulder abduction	Cable row
Lying elbow extension	Lat pulldown
Cable elbow extension	Biceps preacher curl
	Barbell biceps curl

Note: Each exercise of bout routines was performed for 3 sets with a 10 RM load.

Subjects were instructed to maintain their usual nutritional regimen and to avoid taking any supplements during the intervention period. A 24-hour food recall on two nonconsecutive weekdays and one day at the weekend was adopted to assess dietary nutrient intake, which was analyzed through NutWin software (UNIFESP, São Paulo, Brazil). On weeks one (baseline) and five (post-intervention period), subjects were submitted to consultations with a dietitian and a member of the research team in order to ensure that they had not changed their dietary habits during the supplementation period.

Upper- and lower-body maximum strength was assessed by 1RM testing in the bench press ( $1RM_{BENCH}$ ) and parallel back squat ( $1RM_{SQUAT}$ ) exercises with aims of sample characterization. Subjects reported to the laboratory in the first week of the study having refrained from any exercise at least 48 hours before baseline and post-intervention assessments. 1RM testing was consistent with recognized guidelines, as established by the NSCA (15).

Volume load (sets  $\times$  repetitions  $\times$  external load) was calculated for every RT sessions (13). The weekly volume load was calculated as the values corresponding to the sum of the loads calculated for the RT sessions of weeks two and five. The VI was calculated dividing volume load by the body mass of each participant (13).

Subjects reported their session-RPE (sRPE), according to the OMNI-Resistance Exercise Scale validated to measure RPE in RT (17). Subjects were shown the scale ten minutes after each session and asked: "How intense was your session?" and were requested to make certain that their RPE referred to the intensity of the whole session rather than to the most recent exercise intensity. The weekly RPE was calculated as the mean RPE of the four weekly training sessions. In addition, the mean RPE (mRPE) of the four-week intervention period was also calculated. The data were expressed in arbitrary units (AU).

#### *Statistical Analysis*

The normality and homogeneity of the variances were verified using the Shapiro-Wilk and Levene tests, respectively. A paired t-test was used to compare mean values of the descriptive variables, total capsules consumed, total RT sessions performed, VI and RPE. A 2x2 repeated

measures ANOVA (interaction groups [BA and PLA]  $\times$  time [pre- vs post-intervention]) was used to compare the food intake variables (Total caloric intake, grams of carbohydrates/protein/fat, percentage contribution of each macronutrient to total energy intake, macronutrient relative intake). Post hoc comparisons were performed with the Bonferroni test (with correction). The adopted significance was  $p \leq 0.05$ . Furthermore, the magnitude of the differences was examined using the standardized difference based on Cohen's  $d$  ( $d$ ) units by means of effect sizes (8). The  $d$  results were qualitatively interpreted using the following thresholds:  $< 0.2$ : trivial, 0.2-0.6: small, 0.6-1.2: moderate, 1.2-2.0: large, 2.0-4.0: very large, and  $>4.0$ : nearly perfect. Data analysis was performed using a modified statistical Excel spreadsheet (8) and SPSS-22.0 software (IBM Corp., Armonk, NY, USA). The figures were formatted in GraphPad Prism version 6.0 software (La Jolla, CA, USA).

## RESULTS

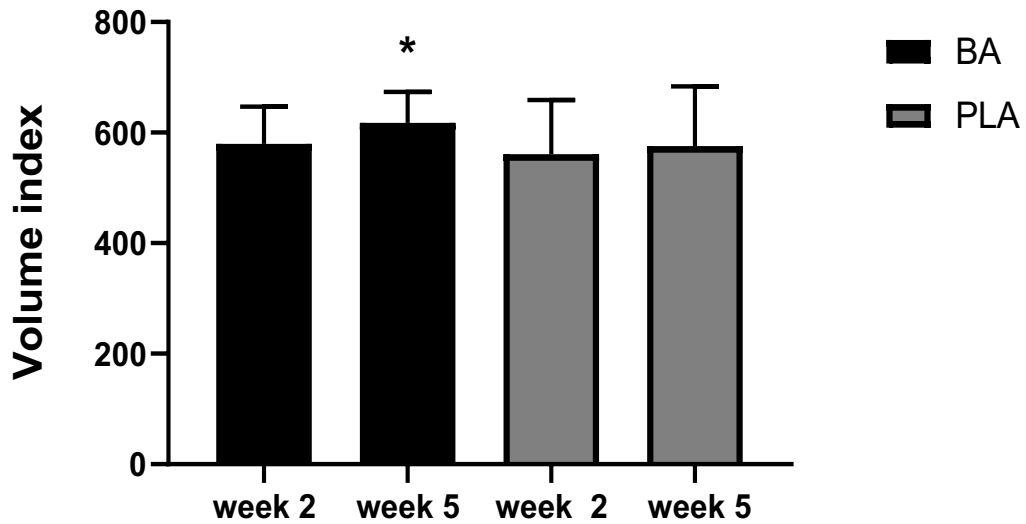
No significant difference was observed between groups for any baseline measurements (all  $p > 0.05$ ) (Table 2). There was no significant difference in any dietary intake variable either within- or between-groups over the course of the study (all  $p > 0.05$ ). No significant difference was noted between groups for total capsules consumed ( $420.0 \pm 23.9$  [ $93.7 \pm 5.5\%$  of compliance] vs.  $424.4 \pm 26.8$  [ $94.6 \pm 5.3\%$  of compliance], BA vs. PLA, respectively) and total training sessions performed ( $14.6 \pm 2.0$  vs  $14.4 \pm 1.0$ , BA vs. PLA, respectively) during the intervention period (both  $p > 0.05$ ). Two participants correctly guessed that they were taking BA, while three correctly guessed that they were ingesting PLA.

**Table 2.** Baseline descriptive statistics (mean  $\pm$  SD).

Variables	PLA ( $n = 8$ )	BA ( $n = 8$ )	<i>P</i> value
Age (years)	26.5 $\pm$ 4.5	28.1 $\pm$ 5.4	0.521
Total Body Mass (kg)	85.4 $\pm$ 10.2	83.2 $\pm$ 5.0	0.085
Height (cm)	178 $\pm$ 0.2	177 $\pm$ 0.1	0.652
RT Experience (years)	6.0 $\pm$ 4.0	5.9 $\pm$ 4.2	0.493
RT Frequency (sessions wk <sup>-1</sup> )	5.0 $\pm$ 0.2	4.8 $\pm$ 0.7	0.653
Energetic Intake (kcal)	2670 $\pm$ 120	2584 $\pm$ 135	0.610
Protein intake (g/kg)	1.5 $\pm$ 0.3	1.6 $\pm$ 0.4	0.375

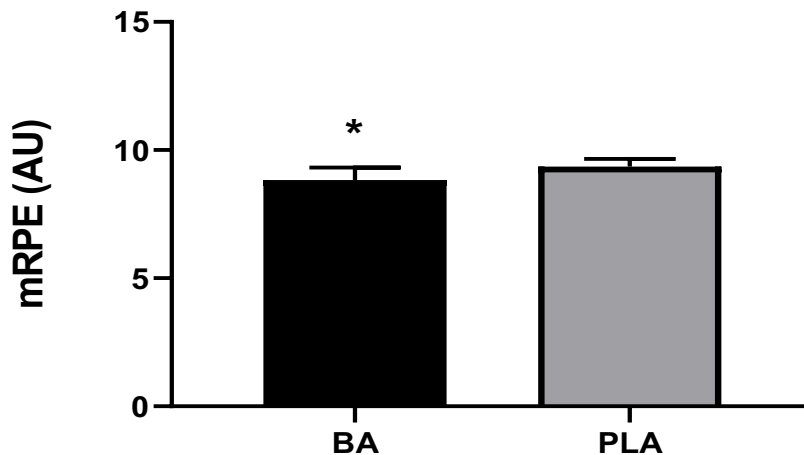
PLA = placebo group, BA = beta-alanine group, RT = resistance training, sessions wk<sup>-1</sup> = sessions per week, kcal = kilocalories, g/kg = grams per kilogram of body mass.

Figure 1 shows the values of VI for each groups during weeks two and five. Significant increases from weeks two to five were observed only for BA group (BA: + 6.5%,  $d = 0.61$ ,  $p = 0.04$ ; PLA: + 2.7%,  $d = 0.14$ ,  $p = 0.11$ ).



**Figure 1.** Volume index for BA (beta-alanine) and PLA (placebo) groups during the second and fifth weeks of the study. \* Significantly different from week two ( $p < 0.05$ ).

For RPE, significant decreases (weeks two to five) were observed only for BA (BA:  $-10.6\%$ ,  $d = 1.96$ ,  $p < 0.001$ ; PLA:  $+1.7\%$ ,  $d = 0.28$ ,  $p = 0.99$ ). Figure 2 presents the values of mRPE for each experimental group. A significant lower value was observed for BA ( $8.8 \pm 0.5$  AU) compared to PLA ( $9.4 \pm 0.3$  AU,  $d = 1.39$ ) ( $p = 0.02$ ).



**Figure 2.** Mean RPE values (mRPE) during the four-week intervention period for beta-alanine (BA) and placebo (PLA) groups. \* Significantly different from PLA ( $p < 0.05$ ).

## DISCUSSION

The aim of the present study was to assess the short-term effects of BA supplementation on the volume load and measures of perceived effort of resistance-trained subjects. Confirming our initial hypothesis, significant increases in VI were observed only for BA group. Additionally, the supplementation protocol induced lower values of RPE.

BA has been reported as a relevant ergogenic aid for high-intensity exercises, especially when higher levels of muscle acidosis are observed (5). Due to the predominant anaerobic character of RT, it can be suggested that increased carnosine levels may lead to significant improvements in performance during the training sessions.

Training volume was expressed as VI in the current study, which considers athlete's body mass in the calculation. This calculation method results in significantly different estimates of workload when compared to the traditional volume load calculations (13). The significant increases in VI observed only for the BA group during the experimental period may be attributed to an increased muscle carnosine content and a higher buffering capacity, since a similar supplementation period with low daily dose (3.2 g) have been shown to be sufficient to increase carnosine content (22). In addition, a 58% increase following 4 weeks of supplementation with the same absolute dosage adopted in the current study has been previously reported (5).

The short-term effect on VI observed in our study is in accordance with previous investigations. However, it is important to note that these other studies reported volume-related outcomes of single exercises (6) or through the number of repetitions performed in a given session (7). In addition, differences regarding the training level of the participants must be considered when comparing these investigations with our study. Therefore, BA supplementation seems to be a viable ergogenic aid to RT practitioners that aim to increase VI. Although speculative, it can be suggested that a longer intervention period would be able to induce larger differences between the experimental groups, since the increases in muscle carnosine have been shown to be larger within longer supplementation protocols (5).

A significant lower rating of perceived effort was observed following BA supplementation. Our results corroborate findings from Hoffman et al. (7), who reported lower feelings of fatigue in the supplemented group. Although different subjective scales and different BA dosages were adopted between the current study and Hoffman et al. (7), a similar percentage difference (5.6% and 6.6%, respectively) in mRPE between BA and PLA groups was observed. Different from the present study, BA supplementation for four weeks failed to reduce RPE values in both Tobias et al. (24) and Roveratti et al. (18). These distinct outcomes might be mainly justified by differences in the training background of the participants of both studies (judo competitors and untrained adults for Tobias et al. (24) and Roveratti et al. (18), respectively). In addition, both studies did not submit the participants to a RT-protocol, which also must be considered in an attempt to compare with our findings. Then, one can assume

that the effects of BA on RPE may be dependent of the training modality and also be modulated by concomitantly performing a RT-program.

The perceived effort/fatigue may be influenced by several factors, such as metabolic and chemical changes that occur during exercise sessions (16). Although speculative, it can be suggested that the acidosis control induced by the ingestion of BA might have been able to reduce the RPE reported by the participants of the present study. The authors of the current study suggest that additional investigations assessing muscle pH along with RPE must be addressed in order to confirm such hypothesis. It is interesting to note that, likewise the present study, the other investigations regarding BA and the perceived effort also adopted short-intervention periods. Thus, it remains to be investigated if longer supplementation protocols would be able to enhance these RPE-outcomes reported.

The present study is not without limitations. First, the duration of the study was quite short, lasting only four weeks. Thus, it is not clear whether these results would have changed if the intervention was carried out over a longer time-frame. Secondly, no methods were used to measure muscle carnosine content, limiting inferences about the real effects of this variable on the dependent variables assessed. Third, the small sample size might have reduced statistical power; thus, this study would be better classified as pilot work and further research is required in order to clarify our findings. It is also interesting to suggest futures studies assessing the effects of the increased volume induced by BA supplementation on morphological outcomes (fat free mass and muscle size). Lastly, these results must not be extrapolated to other populations, such as untrained or high-level competitive lifters.

In conclusion, BA supplementation for four weeks induced larger increases compared to PLA on training volume and a reduced perceived effort in recreationally trained individuals. From a practical standpoint, practitioners aiming to accumulate more training load during a short period of time, such as four weeks, and/or to experience lower levels of effort may benefit from ingesting 6.4 g/day of BA.

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