Effects of Beta-Alanine Supplementation on Athletic Performance: A Mini-Review

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Abstract

Background and purpose: Diet and muscle fiber type are all major factors in predicting intramuscular carnosine concentration [1]. Constituent substrate availability, including the amino acids L-histidine and more specifically beta alanine, play the greatest role in determining the concentration of intramuscular carnosine, a potent intracellular Hydrogen ion buffer. The purpose of this review is to analyze the efficacy of beta alanine supplementation on exercise performance, specifically activities relying on anaerobic glycolysis.

Method: The review included articles from peer-reviewed journals with sufficient data related to the purpose and focus of the study. Inclusion criteria included randomized control trials, systematic reviews and meta-analysis published since 2007.

Results: Twenty relevant studies were identified; various experimental protocols were employed, including both acute and chronic effects of beta alanine supplementation on physical performance. All studies were published 2007 through 2017, providing a robust overview of experimentation over the last 10 years.

Discussion and conclusion: Among studies analyzed in this mini-review, the consensus reached regarding the efficacy of beta alanine supplementation for performance enhancement was relatively positive. Most studies followed similar supplemental strategies, consuming anywhere from 1.6 to 6.4g/day for 2 to 10 weeks. The majority of these studies demonstrated statistically significant increases in intramuscular carnosine content, a physiological parameter that is positively correlated with sprinting and power performance. Many studies presented results in support of enhanced muscular endurance performance subsequent to supplementation relative to the placebo group. Researchers also noted a decline in peak aerobic capacity concurrent to a delay in the onset of blood lactate accumulation, supporting the notion of enhanced glycolytic capacity. More research is necessary to identify optimal dosing strategies for performance optimization across the spectrum of physical activities.

Introduction

Athletes of all calibers use dietary supplements to improve the efficiency of physical training and performance. Beta-alanine (βA), a commonly used ergogenic aid, is produced endogenously in the liver and is ingested in high-protein foods such as chicken and turkey [2]. βA supplementation is used to improve exercise performance by improving the buffering capacity of muscle, thus delaying the onset of neuromuscular fatigue [3].

Aerobic Endurance

Jordan et al. [2] conducted a study to determine if βA delayed Onset of Blood Lactate Accumulation (OBLA), or improved submaximal endurance performance. The participants in this study were 17 recreationally-active men (24.9±4.7 years, 180.6±8.9cm, 79.25±9.0kg). Each participant ran three times per week and had not consumed dietary supplements during the 6 weeks prior to the beginning of the study. After subject selection, the researchers recorded anthropometric data and the results of the incremental treadmill running protocol of each participant. The study was a double-blind, placebo-controlled investigation with eight participants randomly assigned to consume 6.0g/day βA and nine participants randomly assigned to consume 6.0g/day of the placebo (maltodextrin). The participants all consumed 2.0g of the assigned supplement three times a day with meals. The subjects performed pre- and post-supplementation testing at the same time each day after fasting for 2 hours before assessment [2].

The participants performed two physical tests throughout the study. The participants walked for 3 minutes on a treadmill at a fixed speed of 6.4km per hour for aclimation before the incline was increased 2% every 3 minutes, continuing on until the 5th stage. After the 5th stage, the incline was increased by 3% every 3 minutes until the participant reached volitional fatigue. The researchers recorded the oxygen uptake every 30 seconds throughout the test and used this data to determine the VO2max. The participant had to reach two of the following criteria to determine VO2max: plateau in VO2max for the final two stages completed, achieving a respiratory exchange ratio greater than or equal to 1.10, and/or reaching a heart rate within five beats per minute of the age-predicted maximal heart rate. Participants reported a rating of perceived
exertion (RPE) on a 6-20 scale, provided capillary blood lactate and recorded heart rate within the final 30 seconds of each stage [2].

The results of this study indicate that βA supplementation caused a delayed OBLA as demonstrated by increased heart rate at OBLA and percent of maximum heart rate reached at OBLA compared to those statistics in the placebo group results. The percent of heart rate max at OBLA increased pre/post in the βA group (83.0±9.7 to 88.6±3.7, p<0.05) versus no change in the placebo group (86.3±4.8% to 87.9±7.2, p>0.05) [2]. The βA group achieved an increase in body mass (77.9±9.0 to 78.3±9.3kg, p<0.05), where the placebo group did not achieve any change in body mass (80.6±9.1 to 80.4±9.0kg, p>0.05). Other changes in physical performance and characteristics was an increase in percent VO2max at OBLA within the βA group as well as a decrease in VO2max overall for the βA group. The percent of VO2max at OBLA increased in the βA group pre/post (69.1±11.0 to 75.6±0.7ml/kg/min, p<0.05) but remained unchanged in the placebo group (73.3±7.3 to 74.3±3.7ml/kg/min, p>0.05). VO2max decreased in the βA group pre/post (4.57±0.8 to 4.31±0.8L/min⁻¹, p<0.01. The researchers noted that the decrease in VO2max was an unexpected finding. The researchers concluded that the supplementation of βA may have optimized the relative contribution of the anaerobic energy system while reducing the capacity of the aerobic energy system [3].

Carnosine

Beta-alanine supplementation has been shown to cause elevated levels of muscle carnosine content [2]. While the role of carnosine in skeletal muscle is not fully understood, the presence of βA and carnosine has been shown to improve anaerobic exercise performance in untrained subjects. Baguet et al. [4] performed a study examining the relationship between physical performance and muscle carnosine content, as well as if βA supplementation improved anaerobic performance in highly trained rowers [4]. Furthermore, the purpose of this study was to determine if there is a relationship between muscle carnosine content and rowing speed at four different distances. The study was completed in two parts: the first part to determine the relationship between muscle carnosine levels and rowing performance and the second part to determine the effects of βA supplementation. The subjects for this study were 19 elite rowers who volunteered for the study. None of the rowers consumed outside ergogenic supplements within 3 months prior to or during the study. The age, weight, and heights of the participants were recorded prior to testing (23.2±4.4yrs, 84.2±7.8kg, and 188.0±4.4cm, respectively). The training volume of the participants was 10.2±2.9 training sessions per week. In the first part of the study, 15 of the rowers completed timed performances of four different rowing distances (100m, 500m, 2,000m, and 6,000m) and four of the rowers only completed the 2,000m race. The researchers measured the carnosine content in the soleus and gastrocnemius medialis muscles of the lower leg using proton magnetic resonance spectroscopy [4].

For the first part of the study, the researchers tested rowing performance of each participant using the tests of four different distances as previously mentioned. The 100m and 500m tests were completed on Monday of the testing week, the 2,000m test on Wednesday, and the 6,000m test on Friday. All participants completed these tests on a rowing ergometer. Post hoc analysis was performed on the completion times of each participant with each rower ranked from lowest to highest in terms of carnosine content. The rowers were grouped into either a “low carnosine” group or a “high carnosine” group [4]. The second part of the study was a double-blind, placebo-controlled study with 18 volunteer elite Belgian rowers (17 male, 1 female). Eight athletes were randomly assigned to the experimental βA supplementation group and nine were assigned to a placebo group. The participants were supplemented daily for 7 weeks with either 5g/day (divided over five doses of 1g, ingested with 2 hour intervals) βA or an isocaloric placebo (maltodextrin). The participants were tested before the beginning of the supplementation period and in the last week of the supplementation period for muscle carnosine content and rowing performance. The researchers tested for lactate concentration after the warm up, following the 2,000m all out and after 3min of recovery [4].

The researchers found a positive correlation between the mean baseline muscle carnosine concentration in the athletes and the speed of a 100m (P=0.018), 500m (P=0.007), 2,000m (P=0.006), and 6,000m (P=0.003) all-out. The group with high muscle carnosine content was significantly faster during the second and third 500m split of the 2,0000m test, which was typically the slowest part of the race [4]. The second part of the study showed that the carnosine content of the gastrocnemius muscle increased by 28.25% (P<0.013) and by 45.3% (P<0.001) in the soleus muscle in the βA supplementation group and did not change in the placebo group. The carnosine content of the soleus muscle increased between 5% and 71% in the βA supplementation group (p<0.044), indicating a large range of effect. The researchers did not observe a statistically significant difference between performance in the βA group and the placebo group from the beginning to the end of the 7-week study, but there was a significant positive correlation between muscle carnosine content and performance improvement on the 2,000m test (p=0.042). The blood lactate concentration was not found to have changed significantly from the beginning to the end of the study for either group [4].

The results of this study support the conclusion that βA supplementation neither improved physical performance nor improved blood lactate concentrations in rowing athletes. However, the positive correlation between βA supplementation and muscle carnosine concentration may support the theory that βA supplementation has an effect on muscular endurance performance and allow athletes to be faster as muscle carnosine was a strong predictor of speed in each of the four events in this study. The researchers proposed that this relationship may be best explained by the relationship between muscle carnosine concentration and coping with acidosis. Further research should be conducted on the relationship between muscle carnosine concentration, βA supplementation, and acidosis in physical endurance performances [4].
Muscle carnosine is an endogenously produced di-peptide that efficiently buffers hydrogen ions for pH levels in the normal physiological range (7.35 to 7.45). Muscle cells tend to have the highest concentration of hydrogen ions in the body, due to oxidative phosphorylation taking place in the mitochondria, and carnosine helps maintain a healthy level of pH in the muscles for muscle functioning. Thus, muscle carnosine staves off neuromuscular fatigue during exercise. As humans age, neuromuscular fatigue sets more quickly. Researchers conducted a study of physical working capacity at fatigue threshold (PWCFT) to assess the effects of βA supplementation on intracellular pH control and muscle endurance in elderly athletes [5]. This study was conducted using a double-blind placebo controlled design. The participants were comprised of 26 elderly men and women living in independent-living communities who volunteer to participate in the study. The participants were randomly assigned to a βA supplementation group (n=12) or a placebo control group (n=14). The age, body mass, and height characteristics of the βA supplementation group were 72.1±10.6 years, 74.2±16.2kg, and 159.6±9.6cm, respectively. The intervention part of this study was conducted using one capsule of 800mg of Carno Syn βA or, for the placebo group, one capsule of microcrystalline cellulose, ingested three times per day with meals for a total of 90 days. The participants were asked to maintain normal dietary patterns and to refrain from exhaustive exercise, caffeine, and alcohol consumption 24 hours prior to testing [5].

The testing procedures involved electromyography and determination of PWCFT to determine the difference in effects of the placebo compared to the βA supplementation. The researchers used bipolar surface electrodes placed on the right thigh over the lateral portion of the vastus lateralis muscle at the midpoint between the greater trochanter and the lateral condyle of the femur for consistency in placement between participants. PWC_{FT} values were determined using the EMG amplitude values from the vastus lateralis muscle while the participants pedaled on a cycle ergometer. The participants started pedaling at 50rpm and power output was increase 10 to 20W for each 2-minute stage of discontinuous protocol. Between each bout, a rest interval lasted until the heart rate of the participant decreased to within 10rpm of the heart rate measured upon arrival to the laboratory. EMG samples were taken six times for 10 seconds at a time during each 2-minute bout. The PWC_{FT} was determined by averaging the highest power output that resulted in a non-significant (p>0.05; single-tailed t-test) slope value for the EMG amplitude vs. time relationship and with the lowest power output that resulted in a significant (p<0.05) slope value [5].

The results of this study indicated that PWC_{FT} increased significantly (28.6%, p<0.05) from pre- to post-supplementation for the βA supplementation group whereas the results did not indicate a statistically significant increase in PWC_{FT} for the placebo group. The βA supplementation group showed that 67% of those participants improved, but only 21.5% of the placebo group improved. The researchers found that PWCFT testing produced a more reliable test than VO_{2max} because the population tested was not fit enough to endure maximal exertion testing. The researchers concluded that the improved ability to buffer hydrogen ions reduced the rate of fatigue onset and increased capacity for exercise in older men and women [5]. The results of the study indicated that βA supplementation allowed elderly men and women to reach higher PWC_{FT} than a placebo group undergoing the same exercise testing regimen over the course of 90 days. The researchers concluded that this was due to the buffering capacity of intracellular carnosine which was hypothesized to act as an immediate line of defense for the decreases in intracellular pH level that accompany physical exercise. The limiting factor for carnosine synthesis is the availability of βA, thus βA supplementation allows for replenishment of muscle carnosine levels during exercise. Further research should be conducted on the relationship between initial muscle carnosine levels and improvements to carnosine levels with βA supplementation [5].

Unlike elderly athletes, muscle carnosine levels are typically high in trained male sprinters. In theory, muscle carnosine produces greater excitability by increasing Ca2+ sensitivity of skeletal muscle. The effects of βA supplementation show diminishing returns as the natural concentration of endogenously produced carnosine increases, so βA supplementation provides less of a benefit for athletes with naturally higher carnosine levels. Sprint-trained athletes tend to have higher muscle carnosine levels than untrained subjects or endurance-trained athletes. Researchers conducted a study on the effects of βA supplementation on sprint-trained athletes and measured the initial muscle carnosine levels of the athletes as well as the effects of βA supplementation on iso-kinetic and isometric muscle fatigue and post-supplementation βA levels [6].

The participants in this study were 15 male track-and-field athletes with personal records on the 400m sprint below 53 seconds. The average personal record time on the 400m was 50.45±1.60s over a range of 47.49 to 52.64s. The participants were grouped by club and trainer used to minimize the effects of different training amounts and intensities on pre- and post-performance results. The placebo (n=7) group and the βA (n=8) group conducted an average of 5.4 and 5.6 training sessions each week, respectively. None of the participants had consumed oral ergogenic supplements for the 3 months leading up to the study. The body weight, height, and age of the participants were measured prior to the start of the study. The placebo group had weights, heights, and ages of 70.7±5.7kg, 183±4cm, and 18.4±1.5 years, respectively. The βA supplementation group had measured weights, heights, and ages of 74.1±7.2 kg, 184±8cm, and 23.8±4.2 years, respectively [6].

The study was placebo-controlled and double blind. The subjects were supplemented orally for four to five weeks with either a placebo (maltodextrin) or βA (carnosyn) in capsules of 400mg administered six times per day (with at least 2 hours separating each dose). The subjects consumed 2.4g/day of the assigned treatment for the first 4 days, 3.6g/day for the next 4 days, and then 4.8g/day until the end of the study. The participants conducted a familiarization session to get accustomed to the muscle torque measurement procedures of the study a week before pre-
treatment measurements. The researchers measured carnosine concentrations using proton magnetic resonance spectroscopy (MRS) and magnetic resonance imaging (MRI) to scan the entire body of each participant [6].

The participants conducted a standardized warm-up of ascent and descent of stairs over eleven kilometers of altitude and two 50-meter runs at a moderate pace. The participants then performed five sets of 30 repetitions of maximal voluntary iso-kinetic knee extension on the right leg at a constant angular velocity of 180º/s from the position of 90º flexion to full extension. Each participant rested for 1 minute between each bout of 30 repetitions. The peak torque for each contraction was measured to calculate the average peak torque. The participants then conducted isometric tests on the left leg. The knee angle was fixed at 45º and the maximal static voluntary contraction (MVC) torque was determined. The highest torque of three 3-s attempts, separated by 30s of rest, was considered as MVC. Subjects were then asked to contract isometrically with their knee extensors at a target torque of 45% of MVC for as long as possible to determine isometric endurance. The participants completed a 400m sprint for time following a 45 minute warm up. Blood lactate was testing 2-3 minutes before the start of the timed 400m sprint and also at 90s and 180s following completion of the run [6].

The results of the study indicated that βA supplementation increased muscle carnosine levels. The carnosine content of the soleus increased by 47% (P<0.0001) in the βA group to 11.39±1.38mmol/L whereas it remained stable at 7.85±1.04mmol/L (+8%, P=0.41) in the placebo group. In the gastrocnemius, the muscle carnosine concentration increase was significantly more pronounced in the βA group (+37%; P=0.0001) than in the placebo group (+16%; P=0.005). The results also indicated that, despite the expected naturally high levels of carnosine in sprint-trained athletes, the increase in muscle carnosine concentration in the βA group (40% increases over baseline in the gastrocnemius) indicated that there was no ceiling effect. Analysis of the repeated bouts of maximal isokinetic knee extension revealed a significant pre-/post-treatment effect in the first two bouts of 30 repetitions, but not in the subsequent three bouts. The peak knee extension torque of each contraction during the last two bouts was consistently higher after treatment than before treatment. For the 400m sprint, time decreased on average by about 0.7 seconds to 51.44±1.57s in the placebo group and 50.36±1.43s in the βA group. This difference was statistically significant (P=0.02). The blood lactate concentration levels post-treatment increased about 1.2mmol/L in both groups. However, the changes in muscle carnosine content did not significantly correlate with changes in 400m running speed, in maximal lactate accumulation, or with isometric endurance [6].

The results of this study indicate an inconclusive relationship between muscle carnosine levels and changes in running speed, maximal lactate accumulation, and isometric endurance in sprint-trained athletes. However, the results did indicate that βA supplementation was an effective treatment to increase muscle carnosine levels. This study showed that there was no ceiling effect presents preventing additional βA supplementation from increasing already high muscle carnosine levels. The researchers hypothesized that the lack of effect of muscle carnosine on 400m sprint performance was due to intramyocellular buffer capacity not being increased enough by the supplementation to create a significant difference in pre-/post-treatment times. Further research should be conducted on the pH buffering capacity of βA supplementation using proton MRS to noninvasively quantify muscle carnosine content among other variable of interest [6].

βA supplementation has been tested for effects on muscle carnosine levels and resulting increased buffer capacity of hydrogen ions, but it may also affect levels of several hormones related to exercise performance. Testosterone levels increase with higher volumes of resistance training. Growth hormone levels have been shown to fluctuate in response to changes in the acid-base balance of muscle. Researchers conducted a study to determine if βA supplementation had an effect on the acute endocrine response to resistance training. The researchers found that βA supplementation was effective in increasing the gain in performance from a resistance-training workout, but did not have a significant effect on cortisol, growth hormone, testosterone levels, or body mass, 1RM strength or peak power performance [7].

The subjects for this study were comprised of eight resistance-trained (greater than or equal to 3 years of experience) college-aged males. The age, height, body mass, and percent body fat of each participant was measured and recorded for each participant for averages of 19.7±1.5 years, 176.8±3.7cm, 89.0±7.9kg, and 15.7±2.8% body fat, respectively. The subjects were not permitted to use any other nutritional supplements or anabolic steroids during the conduction of the experiment [7]. The study was performed as a double-blind, randomized cross-over design. The subjects conducted pre-treatment testing consisting of a 1RM of a free weight squat. The first testing session then consisted of conducting six sets of twelve repetitions of 70% of that 1RM weight from the pre-treatment testing with 1.5 minutes of rest between each set. The subjects then began a 4 week long βA or placebo supplementation period. Following the supplementation period, the subjects returned to conduct another 1RM testing of the free weight squat exercise. Then three days after the post-treatment 1RM test, the researchers conducted testing for final acute resistance exercise post-bout for endocrine response levels. The subjects stopped using the supplements for another 4 weeks for a washout period then returned for another 1RM test. Following the washout period, the subjects began another 4 week long supplementation period receiving the opposite supplement form the previous 4 week period. At the end of the second treatment period, the subjects conducted another 1RM test and final acute resistance exercise testing session [7].

The 1RM free weight squat tests were performed after each participant completed two warm up procedures. Each participant conducted a regular warm up protocol for 5 minutes and then conducted 2 to 3 minutes of light to moderate weight (40-60% of 1RM) squats. For testing, each participant was instructed to squat...
to the parallel position under control, but return to the starting position (full leg extension) as quickly as possible. Repetitions not completed to this standard were discarded for data analysis.

All experimental testing occurred at the same time of day to reduced effects of diurnal variation. Power was measured for each participant by attaching a measuring device to the end of the barbell to measure linear displacement and time. Bar velocity was calculated and power was determined when bar load was manually input. Baseline blood samples were obtained before each experimental testing session, as well as immediately following post-exercise (IP), 15 minutes after exercise (15P), and 30 minutes after exercise (30P). The researchers used enzyme immunoassay (EIA) to determine serum testosterone, growth hormone, and cortisol levels of the blood samples. The researchers noted that there was no significant change in testosterone in any trial (p=0.08) [8].

The results of the study indicated that the subjects increased training volume during βA supplementation, but that βA supplementation did not have a significant effect on cortisol, testosterone, or growth hormone levels. Significant differences in training volume between βA and PL groups (193±834kg and 145±1491kg, respectively) as well as total number of repetitions performed during the workout (9.0±4.1 and 0.3±7.8, respectively) were observed. There was also a significant difference in change in mean power between βA (98.4±43.8W, p≤0.05) and PL (7.2±29.6W, ps≤0.05) groups. Both groups demonstrated increase lactate concentrations. However, the tests conducted after the washout period indicated that the group which had previously consumed βA still exhibited increased training volume capability, indicating that the washout period may have been too short to accurately assess the differences between βA supplementation and placebo effect [9].

The researchers concluded that the βA supplementation regimen in this study improved response to resistance training sessions. However, the improved response was unrelated to changes in testosterone, cortisol, or human growth hormone levels. While the performance tests indicated a significant difference between the βA supplementation and placebo groups, there was no significant difference between the second iteration βA supplementation group and the second iteration placebo group. The researchers hypothesized that this lack of significance was due to the brevity of the washout period. If the washout period was too short, then the first βA supplementation group was still being affected by the supplementation before the washout period [7].

Power

Female soccer players frequently conduct plyometric training and supplement with beta-alanine [10]. Researchers conducted a study to compare the effects of plyometric training with and without βA supplementation on endurance and maximal-intensity exercise testing. The researchers had previously found that the plyometric training program improved performance in several physical tests (explosive jumping, sprinting, repeated sprinting, 60s repeated jumping, endurance, and change-of-direction speed performance) and hypothesized that βA supplementation may further enhance the effects of the plyometric program [10]. The subjects for this experiment consisted of 25 amateur female soccer players who were not otherwise involved in regular strength or plyometric training within the 3 months prior to the study and had never take βA supplementation prior to the study. Participants were assigned to either a plyometric training group and placebo supplementation, a plyometric training group and βA supplementation, or a control group with a placebo or no plyometric training protocol. Body height, body mass, squat jump, countermovement jump, 20m sprint test, running anaerobic sprint test, 20cm drop jump reactive strength index, peak power jump change of direction speed, 20m multistage shuttle run, 60 second countermovement jump of each subject were measured one week before and one week after the intervention [10].

The results of the experiment showed that the players did not experience a change in body mass in any of the groups (control 58.5±7.2kg, placebo 61.1±8.3kg, and βA 58.1±6.3kg, p=0.05). Both groups doing plyometric training demonstrated increased squat and countermovement jumps, drop jump reactive strength indices, jump power performances and achieved a greater increase in each test compared to the control group. There was no difference between the βA supplementation group and the placebo groups (six equal doses of 0.8g/dose consumed every 2 hours each day for six weeks of intervention) in improvements for jumping and power performance (p<0.05; ES=0.27 to 1.0) in each test except for the 60s countermovement jump power test in which the βA supplementation group showed greater improvements. The running anaerobic sprint test (RAST) test, change of direction speed, 20m sprint and 20m multistage shuttle run tests showed improvements (p<0.05) for both plyometric training groups with the βA supplementation group showing the greater improvement on the RAST and 20m multistage shuttle tests compared to the control group [10].

The real-world application of βA supplementation on athletic performance would be attractive to athletes at all levels as long as the benefits of βA supplementation actually improve athletic performance and scoring in competitive sport. Iso-kinetic and isometric physical tests provide clear data for researchers to interpret, but studies typically do not extend from simple measurements to improvements in game play. Researchers designed a study to assess physical performance on the Loughborough Intermittent Shuttle Test (LIST), an exercise which mimics the alternation between low and high-intensity running during competitive games, and the effects of βA supplementation on LIST performance. This study assessed changes in performance in both elite and non-elite athletes [11]. The subjects for this study were comprised of 16 elite and 20 non-elite male athletes. The elite players were randomly assigned to either a placebo group (n=8) or a βA supplementation group (n=8), and the non-elite players were also randomly assigned to either a placebo group (n=10) or a βA supplementation group (n=10). The researchers recorded the average ages, heights, body masses, estimated VO2max and compliance for each group. The measurements of the elite PL group were 19±2 years, 1.77±0.05 m, 72.1±7.1kg, 59±2.6 VO2max and 94±5% compliance. The measurements of the elite βA
supplementation group were 20±1 years, 1.80±0.06m, 75.0±11.0kg, 58.6±2.4ml/kg/min estimated VO2max, and 87±10% compliance. The measurements of the non-elite PL group were 22±3 years, 1.81±0.07m, 84.9±10.9kg, 50.7±5.0ml/kg/min estimated VO2max, and 96±4% compliance. The measurements of the non-elite βA supplementation group were 22±2 years, 1.79±0.80m, 81.0±11.5kg, 50.5±4.4ml/kg/min estimated VO2max and 96±6% compliance. The participants had not taken any supplements during the 3 months preceding the study and had not used βA supplements within the 6 months preceding the experiment [11].

The physical testing consisted of two session of a multistage fitness test and two sessions of List testing. The participants first warmed up with a 5 minute standardized warm up consisting of light jogging and running followed by 5 minutes of self-selected stretching. The first testing exercise was a progressive shuttle run test to exhaustion. The participants had to run between markers set 20m apart at increasing speeds dictated by an audio signal. The participants continued running between the markers until failure to reach the designated line two consecutive times or at volitional exhaustion. The final level attained by the participant was used to estimate maximal oxygen uptake. This portion of the experimental design constituted the familiarization and initial testing. The main testing procedures consisted of List testing. List consists of six exercise sets approximately 15min long separated by periods of 3min rest. Each set consisted of an exercise pattern incorporating walking, sprinting (over 15m) and recovery, cruising and jogging. Cruising and jogging were defined as 95 and 55% of an individual’s estimated VO2max respectively. This exercise pattern was repeated 11 times within each exercise set. Individual sprint times over 15m were recorded for each of the 11 sprints per set, and a total of 66 sprints were completed during the LIST. The sprint times for each set were averaged. The heart rate of each participant was measured every 5 seconds throughout exercise. The participants provided rates of perceived exertion during the last walking stage of each set. The researchers tested blood lactate levels in each participant during the 3 minute rest periods between each set immediately following the final sprint [11].

For the elite groups, the results indicated that there was no effect of supplementation on sprint performance across each set of the List (p=0.9) or over all the sprints in the List (p=0.92). Blood lactate levels did not indicate an effect of supplementation with respect to time (p≤0.001) or heart rate (p=0.76). For the non-elite groups, there was no effect of supplementation on sprint times across each set (p=0.99) or over all the sprints in the List (p=0.58). However there was no effect of time in non-elites (p=0.12) indicating that the sprint times did not significantly decrease as the number of sprints performed increased. Blood lactate levels showed no effect of supplementation (p≤0.001) [11]. βA supplementation did not have a significant effect on sprint performance during the List, however, the researchers caveat that this conclusion may be due to the physical difficulty of the test not being high enough. The participants could all maintain 40 exercise bouts of 15m sprint performance when separated by 30 seconds of rest. The List did not prove sufficiently challenging to induce fatigue. The researchers hypothesize that the βA supplementation may have improved the buffering capacity of the participants and that this results was masked by the insufficient challenge of the test. Further research should conducted using a test similar to the List but challenging enough to push participants past the point of fatigue [11].

**Acute vs. Chronic Effects**

The effects of βA supplementation may or may not be seen in acute supplementation as well as long-term supplementation. Kresta et al. [12] conducted a study examining both the short-term and chronic effects of βA supplementation alone and in conjunction with creatine monohydrate [12]. This study addressed the effects of these two supplements on body composition, aerobic and anaerobic exercise performance, muscle carnosine, and creatine levels in college-ages recreationally active females. The researchers found that there was no evidence of additive benefits of βA supplementation and creatine supplementation in the participants of this study [12]. This study was a randomized double-blind, placebo-controlled trial which included testing at both an acute (7 days) measurement and a chronic measurement (28 days). The subjects selected for this study were healthy, moderately active females between the ages of 18 and 35 years. Each participant had a consistent history of exercising for 3 days a week for 30 minutes at a time for at least 3 months prior to the start of the study. The participants also had to have a history clear of any use of ergogenic nutritional supplementation for at least 3 months prior to the study. The participants performed resting and exercise testing prior to any supplementation, after one week of supplementation, and after 4 weeks of supplementation. The supplementation schedule consisted of a loading phase for the creatine for week 1 of 0.3g/kg of body weight and a maintenance phase for weeks 2-4 of 0.1g/kg of body weight. A βA group received a continuous dose of βA of 0.1g/kg of body weight [12].

The researchers performed percutaneous muscle biopsy on the vastus lateralis muscle of the right leg. The participants fasted for 8 hours prior to testing and all consumed the same meal replacement drink 4 hours before reporting to the lab. The first physical test performed by the participants was a maximal graded exercise test (GXT) completed on a cycle ergometer. The participants began at 50W and increased 25W every 3 minutes until a pedaling rate of 70rpm could no longer be maintained. The researchers tested capillary lactate levels in the final minute of each stage of exercise and five minutes into the recovery period. After 30 minutes of rest, participants also completed two 30 second Wingate Anaerobic capacity test (WAnT) with a standardized work rate of 7.5Kg/rev. The capillary lactate levels of each participant were measured before the start of the first Wingate test, immediately following the second Wingate test, and after 5 minutes of passive recovery [12]. The results of this study demonstrated that the beta-alanine and beta-alanine plus creatine monohydrate group exhibited the highest levels of increased muscle carnosine levels. However, this relationship was not significantly different from the results of the other groups. The results also showed that there was significant...
time by group effects on body weight, fat mass, fat free mass, and body fat [12]. There were no meaningful changes over time for aerobic capacity performance. Participants in the beta-alanine group exhibited much less change in resting to maximal lactate levels. Future research ought to be conducted on whether the levels of beta-alanine and creatine supplementation were adequate for women as opposed to men [12].

HIIT

The majority of studies focus on the effects of βA supplementation on the muscle carnosine levels or the hydrogen buffering capabilities of participants. Generally, these studies examine performance without the supplement and then after consuming the supplement for a set amount of time and then retesting the blood chemistry and performance levels. However, Smith et al. [13] conducted a study examining both the effect of high-intensity interval training (HIIT) and HIIT combined with βA supplementation over the course of 3 weeks and 6 weeks [13]. The participants in this study were 46 men (22.2±2.7 years, 178.1±7.4cm, 78.7±11.9kg, VO₂ peak of 3.3±0.59l.min⁻¹). The study was double-blind and randomized and used two 3 week periods of HIIT and βA supplementation. The participants conducted pre-, mid-, and post-testing consisting of cycling body composition, and VO₂peak power output. The first 3 week of training involved HIIT with workloads of 90-110% of VO₂peak power output of each participant. The experimental group supplemented with 6g of βA per day while the control group consumed a placebo for the first 3 weeks of testing. After the first 3 weeks, the experimental group reduced supplementation to 3g of βA per day and the control group continued to consume the placebo through the end of the 6 week study [13].

The training intervention of the study was conducted using an electronically braked cycle ergometer. The participants began training at 90% of their individual maximum power output achieved during baseline testing. The training plan utilized undulating periodization. During the first 3 weeks the participants conducted five sets of 2 minute intervals with 1 minute of rest in between. The second 3 weeks of the training intervention increased the repetitions to six repetitions during the last two weeks. The supplements were mixed with orange flavored drink powder and water before consumption and the beverage was consumed in two doses, the first 30 minutes before the exercises and the second immediately following the exercises [13]. The results of this study showed that HIIT alone was able to produce a statistically significant increase in VO₂peak, VO₂ at time to exhaustion (VO₂TTE), and total work done (TWD) after the first 3 weeks of training (p<0.05). Both groups showed an increase in VO₂peak after 3 weeks of training, and there were no significant differences between the two groups for VO₂peak at any point over the 6 weeks (p=0.001). However, the experimental group did demonstrate significant increase in VO₂peak from mid to post-training (p=0.010). There was no significant difference between the two groups in improvement of time to exhaustion, but the improvements were statistically significant compared to the baseline testing values (p<0.05). There was a statistically significant increase in lean body mass for pre- to post-testing only in the experimental group (p=0.011). Both groups demonstrated a 50-53% increase in TWD during the first 3 weeks and the experimental group showed a 32% increase in TWD during the second 3 weeks (compared to the 18% increase demonstrated by the control group) [13].

This study demonstrated the benefits of HIIT training and HIIT training in combination with βA supplementation. The supplemented group demonstrated greater improvements in VO₂peak, VO₂ at time to exhaustion (VO₂TTE), and total work done (TWD) than the control group which performed HIIT without supplements. The control group did not see the same improvements to these variables that the experimental group did during the second 3 weeks of the experiment, however. In order to improve this study, the researchers might form a third testing group conducting HIIT while supplementing with both βA and creatine [13].

Gender

The vast majority of studies examining the effects of βA supplementation utilize all-male testing populations. Neuromuscular fatigue can be detected by the overall increase in electrical activity in a given muscle over time during exercise. Neuromuscular fatigue sets in due to reduced buffering capacity of hydrogen ions that accumulate during physical exercise. Physical working capacity at fatigue threshold (PWC₀₂), ventilatory threshold (VT), and time-to-exhaustion (TTE) may be used to measure the effects of βA supplementation on athletes. Researchers conducted a study using these three techniques in order to determine the effects of βA supplementation in female athletes on PWC₀₂, VT, and TTE [14]. The participants in this study were comprised of 22 female volunteers. The study was conducted as a double-blind, randomized, placebo-controlled, parallel design study. The participants were assigned randomly into either a placebo (PL) group (n=11) or a βA supplementation group (n=11). The average ages, heights, and body masses of the PL group (28.9±8.1 years, 161.1±6.4cm, 28.0±10.5kg, respectively) were recorded at the start of the study. The average ages, heights, and body masses of the βA group (25.8±4.0 years, 164.4±6.7cm, 62.2±10.1kg, respectively) were recorded at the start of the study. The dosage of supplementation consisted of four divided doses for both groups. For the first 7 days, the βA and placebo groups consumed 3.2g/day, respectively. For days 8-28, the dosage was increased to 6.4g/day of the respective supplements for each group [14].

The exercise testing protocol consisted of a continuous graded exercise test (GXT) on an electronically braked cycle ergometer to determine VO₂peak, ventilatory threshold, physical working capacity at fatigue threshold, and time to exhaustion. The participants conducted the physical testing at the same time of day for each subject and used the same equipment. The participants fasted for 3 hours prior to physical testing. The initial power output for the GXT was set at 40W and increased by 20W every 3 minutes until the participant could no longer maintain the required power output at a pedaling rate of 70rpm or volitional termination due
to fatigue. During the physical tests, the researchers measured the respiratory gases of each participant and used these measurement to calculate the minute ventilation ($V_e$), oxygen consumption rate ($V_O_2$), carbon dioxide expiration rate ($V_CO_2$), and respiratory exchange ratio (RER). The highest $V_O_2$ recorded during the GXT was recorded as the $V_O_2max$ the participant demonstrated two or more of the following criteria: a plateau in heart rate, heart rate values within 10% of the age-predicted maximal heart rate, a plateau in $V_O_2$ (increases by no more than 150 ml/min), or a RER value greater than 1.15. Ventilatory threshold was determined using a plot of $V_e$ against $V_O_2max$ to calculate two lines of regression. The $V_O_2max$ associated with the VT break point was used as the representative value [14].

The researchers utilized electromyographic measurements to determine the PWC$_{max}$. The EMG sensors were placed over the vastus lateralis muscle. A reference electrode was placed over the iliac crest. The PWC$_{max}$ was determined by averaging the highest power output from the GXT that resulted in a non significant (p>0.05; single-tailed t-test) slope value for EMG amplitude vs. time relationship with the lowest power output that resulted in a significant (p<0.01) slope value. The participants also provided dietary logs of for the first and fourth weeks of testing which were used to compile total kilocalorie and macronutrient intake [14]. The results showed a 13.9% increase in VT from pre- to post-treatment (p<0.001) for the βA supplementation group. The PL group did not show a change in VT (p>0.05). There was a 12.6% increase in PWC$_{max}$ power output form pre- to post-treatment (p<0.001), and no changes in the PL group (p>0.05). The results indicated no two-way interaction for $V_O_2max$. The results indicated no two-way interaction for TTE (p>0.05) in the βA supplementation group. However, there was a significant change in TTE from pre- to post-treatment (2.5%). There were no significant changes in body mass observed from pre- to post-treatment in either group [14].

The researchers concluded that βA supplementation did delay the onset of neuromuscular fatigue during incremental cycle ergometry in young women as indicated by the increased PWC$_{max}$ (12.6%), VT (13.9%) and TTE (2.5%). The researchers found that these results were similar to the results of other studies on the effects of βA supplementation on female subjects. The researcher determined that the inconsistent findings on TTE may have been caused by any combination of the short length of the supplementation intervention, the dosage, concurrent exercise training by the participants, or the training status of the participants. Further research should be conducted to determine the interactions among training status, concurrent exercise training, βA supplementation in women, and optimal dosage of βA supplementation [14]. The majority of research conducted on βA supplementation uses cyclists, runners, and rowers as the test subjects. Likewise, the majority of research is conducted on male athletes or single-sex teams. Chung et al. conducted a study on the effects βA supplementation on the performance of elite and sub-elite male and female swimmers in training and in competition [15].

The researchers divided the 42 participants (34 male and 26 female) into 3 training groups: Sprint 50m-100m specialists, Middle 100m-200m specialists, or Distance 200m-400m specialists [15]. The swimmers were supplemented with either 4.8g/day of βA during the loading phase and 3.2g/day during the maintenance phase or a placebo for 10 weeks. The researchers tested capillary blood for pH, bicarbonate, and lactate concentration both during competition and training. The researchers measured the sum of the mean times of combined 50m swims and the all-out effort at the end of each training set. The sprint group conducted 4x50m maximal bouts on a three minute cycle and a 100m maximal effort at the end of the training set. The middle distance group conducted 6x50m maximal bouts on a two minute cycle and a 200m maximal effort at the end of the training session. The distance group conducted 8x50m on a one-and-a-half minute cycle and a 200m maximal effort at the end of the training session. The results showed an unclear effect of βA on race performance compared to the placebo. The log transformed performance times for the βA group at baseline and post-supplementation were 454.6±52.5s and 454.6±52.6s, respectively. The placebo baseline and post-supplementation times were 479.0±70.4s and 7478.6±70.7s, respectively. No substantial effects on blood chemistry were found. However, there was a temporary substantial effect of βA after the 4 week loading phase. The researchers found that, even though 59% of the participants correctly guess whether they had been given βA or a placebo, the participants who guessed that they had taken the placebo did not perform better than those who believed they had taken the placebo [15].

The investigators were limited in this experiment by the lack of ability to measure changes in muscle carnosine content through either direct or indirect methods [15]. The lack of significant effect of βA on the performance of the participants in this study may be due to any or all of several factors. Swimming is a sport with inherent variability which is highly susceptible to the influence of the diet of the athlete, the athlete’s mood state, or cumulative fatigue. Additionally, the dosage of βA used in the testing group may have been too small to elicit a substantial enough increase in muscle carnosine to affect performance long term. This study would be improved by the measurement of baseline and post-treatment muscle carnosine levels, an additional testing group supplemented with a higher dose of βA, and MRI scans to determine any growth in muscle size [15]. While fewer studies have been conducted on female athletes, the lower baseline muscle carnosine level in women may offer more room for beneficial effects of βA supplementation. Men express a 3.5:1.0 ratio of muscle carnosine levels to women and βA supplementation has been found to increase muscle carnosine levels similarly to male test subjects, particularly in trained female athletes as trained muscles are more sensitive to βA supplementation. Glenn et al. conducted a study on the effects of acute beta-alanine supplementation on trained female cyclists. Specifically, this study investigates the effects of βA supplementation on anaerobic performance [16].

The participants of this study were comprised of 12 trained, competitively active female cyclists (26.6±1.3years, 161.08±1.78cm, 58.67±1.74kg). All of the testing conducted in this study occurred during the luteal phase of menstruation for each participant. The
participants fasted for at least 6 hours prior to testing in order to control for βA levels due to diet. The participants completed the treatment conditions in double-blind, randomized order with either a placebo of 34g of dextrose or a treatment of 1.6g βA and 34g of dextrose. The participants conducted three maximal effort 30 second Wingate tests each followed by 2 minutes of recovery in between. The researchers tested for blood lactate levels (mmol/L) and heart rate (bpm) before the Wingate test, immediately following the test, and then again following the rest period. The participants reported RPE after each maximal bout and after each rest period [16]. The results of this study indicated that βA supplementation did not alter physical performance as demonstrated by either the fatigue index or power output. βA supplementation did, however, decreased reported feelings of exertion after the first two Wingate tests and after each rest interval (p<0.001). Additionally, βA supplementation was not found to have an effect on blood lactate (p<0.001) or time of increased heart rate (p<0.001). The researchers proposed that these results are in line with those of other studies in that the βA resulted in reduced RPE but not an increase in actual performance [16]. While βA supplementation did not result in increased anaerobic performance in young female cyclists, it did result in reduced RPE following maximal exercise and rest cycles. A reduction in RPE may be valuable despite the lack of increased performance as it is one of the many psychological factors associated with performance. This study would be improved by utilizing timed trials in order to evaluate pacing strategies which might benefit more from a reduction in RPE than maximal effort, fixed time-period tests [16].

Conclusion

While several studies have reported that βA supplementation did not result in significantly different performances in various physical tasks, there were many which did demonstrate a significant difference in performance or measured muscle carnosine levels [10-14]. Smith et al. [13] found that HIIT either alone, βA supplementation alone or both treatments combined resulted in statistically difference performance levels in HIIT tests after several weeks of testing. Further research is necessary to better understand the potential benefits that beta-alanine may play in enhancing physical performance across a broad spectrum of athletic and non-athletic populations alike. Implementing better control factors through stricter subject selection, classifying subjects based upon current intramuscular carnosine levels, and regulating dietary composition are all factors that may play a major role in altering potential future outcomes.

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