Metabolic consequences of β -alanine supplementation during exhaustive supramaximal cycling and 4000-m time trial performance

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ABSTRACT

The present study investigated the effects of β -alanine supplementation on the resultant blood acidosis, lactate accumulation and energy provision during supramaximal intensity cycling, as well as the aerobic and anaerobic contribution to power output during a 4000-m cycling TT. Seventeen trained cyclists ($\text{VO}_{2\text{max}} = 4.47 \pm 0.55 \text{ L} \cdot \text{min}^{-1}$) were administered 6.4 g of β -alanine ($n = 9$) or placebo ($n = 8$) daily for 4 wk. Participants performed a supramaximal cycling test to exhaustion (equivalent to 120% $\text{VO}_{2\text{max}}$) before (PreExh) and after (PostExh) the 4-wk supplementation period, as well as an additional post-supplementation supramaximal cycling test identical in duration and power output to PreExh (PostMatch). Anaerobic capacity was quantified and blood pH, lactate and bicarbonate concentrations were measured pre-, immediately post-, and 5 min post-exercise. Subjects also performed a 4000-m cycling TT before and after supplementation while the aerobic and anaerobic contributions to power output were quantified. β -alanine supplementation increased time to exhaustion ($+12.8 \pm 8.2 \text{ s}$; $P = 0.041$) and anaerobic capacity ($+1.1 \pm 0.7 \text{ kJ}$; $P = 0.048$) in PostExh compared to PreExh. Performance time in the 4000-m TT was reduced following β -alanine supplementation ($-6.3 \pm 4.6 \text{ s}$; $P = 0.034$) and the mean anaerobic power output was *likely* to be greater ($+6.2 \pm 4.5 \text{ W}$; $P = 0.035$). β -alanine supplementation increased time to exhaustion concomitant with an augmented anaerobic capacity during supramaximal intensity cycling which was also mirrored by a meaningful increase in the anaerobic contribution to power output during a 4000-m cycling TT, resulting in an enhanced overall performance. **Key words:** Ergogenic aid; supplement; buffer capacity; carnosine; cycling; time trial.

Introduction:

Carnosine (β -alanyl-L-histidine) is an intramuscular dipeptide found in high concentrations in skeletal muscle and is thought to play an important role in regulating muscle function (Sale et al. 2013). Chronic β -alanine supplementation is the most efficacious nutritional strategy that acts to raise carnosine concentrations in human muscle (Stellingwerff et al. 2012). As such, the ergogenic potential of β -alanine supplementation has been studied across a wide range of exercise performance protocols (Bellinger 2014; Blancquaert et al. 2015). However, data regarding the metabolic and physiological effects of β -alanine supplementation during exercise *in vivo* is less prevalent in the literature.

The chronic supplementation of β -alanine has previously been demonstrated to improve supramaximal cycling time to exhaustion (TTE) (Hill et al. 2007; Sale et al. 2011) explained via an increase in intracellular buffering capacity (Abe 2000; Baguet et al. 2010b). Accordingly, β -alanine supplementation could be responsible for a delay in the accumulation of fatigue-related metabolites (such as hydrogen ions) during supramaximal exercise, and consequently result in an increased capacity for sustained high rates of glycolytic flux. Enhanced anaerobic ATP production following high-intensity interval training has been previously associated with increased TTE during supramaximal exercise (Weber and Schneider 2002). However, it has not been previously determined if β -alanine supplementation is associated with an increase in anaerobic ATP production during exhaustive supramaximal exercise. Considering the significant correlation between anaerobic capacity and performance during short-distance cycling (Craig et al. 1993) and running time trial (TT) (Ramsbottom et al. 1994), the maximum amount of ATP supplied anaerobically may be an important determinant of performance, and may explain, at least in part, the ergogenic potential of β -alanine supplementation on exercise performance and metabolism.

Similar to TTE tests, worthwhile improvements in short-distance rowing (Hobson et al. 2013) and swimming TTs (De Salles Painelli et al. 2013) have also been reported following β -alanine supplementation. For example, De Salles Painelli et al. (2013) reported that β -alanine supplementation improved 100- and 200-m swimming TT performance by 2.1% ($P = 0.029$) and 2.0% ($P = 0.0008$), respectively. Furthermore, Hobson et al. (2013) reported that β -alanine supplementation was *very likely* to be beneficial to 2000-m rowing performance (6.4 ± 8.1 s reduction in time compared with placebo). However, the aerobic and anaerobic contributions to energy provision were not measured in these studies. To date, it is still not known if the improved short-distance TT performance resulting from supplementation with β -alanine is concomitant with an altered metabolism during exercise. Increased post-exercise blood $[\text{La}^-]$ following β -alanine supplementation reported in previous studies was attributed to an increase in glycogen utilization (Donovan et al. 2012; Tobias et al. 2013).

Gross et al. (2014) reported that β -alanine supplementation increased the aerobic energy contribution (1.4 ± 1.3 %, $d = 0.5$), concurrent with a reduced accumulated oxygen deficit (-5.0 ± 5.0 %, $d = 0.6$) and muscle lactate concentration ($[\text{La}^-]$) (-23 ± 30 %, $d = 0.9$), while having no effect on pH. However, in the study by Gross et al. (2014) the authors chose to employ a single exercise protocol of fixed intensity and did not include an additional exhaustive performance test, which has been suggested to be a salient method to compare markers of exercise metabolism and ion regulation (Harmer et al. 2000). Moreover, the accumulated oxygen deficit method (Medbo et al. 1988) employed to quantify anaerobic capacity has been criticized for having limitations as a valid and reliable measure of anaerobic capacity (Noordhof et al. 2010). We speculate that the appropriate analysis of the physiological responses under conditions of identical work, and exhaustive exercise, before and after β -alanine supplementation will resolve these inconsistencies in the literature. Therefore, the aim of this study was to investigate the effects of β -alanine supplementation on

the resultant blood acidosis, lactate accumulation and energy provision during work-matched and exhaustive supramaximal intensity cycling, as well as the contribution of the aerobic and anaerobic energy systems during a 4000-m cycling TT.

Materials & methods:

Participants

Seventeen trained male cyclists (mean $\pm$ SD: age = 24.5 ± 6.2 yr, mass = 70.1 ± 3.7 kg, $\text{VO}_{2\text{max}} = 4.47 \pm 0.55 \text{ L}\cdot\text{min}^{-1}$) were recruited for the current study. At the time of investigation, they were cycling $300\text{--}600 \text{ km}\cdot\text{wk}^{-1}$ and competing in local A grade criterion racing ($n = 13$) and the national road series ($n = 4$). All cyclists were informed verbally and in writing as to the requirements of the study, and all gave their written informed consent. The study was conducted in the Griffith University Sport Science laboratory and was approved by the Griffith University Human Research Ethics Committee (protocol number RHS/50/13HREC). Cyclists were naive to chronic β -alanine supplementation before the commencement of the study and had not taken any nutritional supplements in the 3 mo before the study with the exception of five cyclists who were consuming a multi-vitamin supplement and five cyclists who were consuming a fish-oil supplement during the duration of the study. It should be noted that ten cyclists included in this study acted as subjects in a previous study in our laboratory (Bellinger and Minahan 2015).

Experimental design

In the two weeks prior to supplementation, subjects visited the lab on five occasions (>48 hrs apart). On their first visit to the laboratory, subjects performed a long-graded exercise test to exhaustion for determination of $\text{VO}_{2\text{max}}$. The test started at 100 W and increased by 50 W every 5 min until volitional exhaustion. Maximal aerobic power output was calculated from

the last completed work rate, plus the fraction of time spent in the final non-completed work rate multiplied by the work rate increment. VO_2 was considered to be maximal when at least two of the following four criteria were met: 1) a levelling off of VO_2 with increasing work rate (increase of no more than $2 \text{ mL} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}$, 2) a heart rate within $10 \text{ beat} \cdot \text{min}^{-1}$ of age predicted maximum, 3) $\text{RER} > 1.05$. During the second visit to the laboratory, subjects performed a familiarisation of the supramaximal cycling test to exhaustion (equivalent to $120\% \text{VO}_{2\text{max}}$) for determination of TTE and anaerobic capacity. During their third visit to the laboratory, subjects performed a familiarisation of the 4000-m cycling TT to evaluate performance time, mean power output and the aerobic and anaerobic contributions to energy provision. During visits four and five, subjects performed the pre-supplementation supramaximal cycling test (PreExh) and 4000-m cycling TT, respectively. Following pre-supplementation testing, subjects were matched for training load in the 2 wk before familiarisation testing and randomly assigned to receive either $6.4 \text{ g} \cdot \text{day}^{-1}$ of β -alanine (Carnosyn® slow-release, Collegiate Sport Nutrition, San Marcos, California, USA, $n = 9$) or a placebo (dextrose monohydrate, $n = 8$) ingested in four split doses over each day with meals for 28 days. Following the supplementation period, subjects completed a supramaximal cycling test to exhaustion (PostExh) and a 4000-m TT, as well as an additional supramaximal cycling test which was stopped at the time of exhaustion (PostMatch) that was achieved during the pre-supplementation supramaximal cycling test.

Details of exercise performance tests

Testing was conducted on an electromagnetically braked cycle ergometer (Velotron Racermate, Seattle, WA), which has previously been shown to provide a high level of reliability during dynamic cycling TT performance (Sporer and McKenzie 2007; Abbiss et al. 2008). The ergometer was adjusted for comfort for each subject; this included fitting their own pedals and saddle. These adjustments were replicated for all subsequent trials. Subjects

completed a familiarization of each performance test (supramaximal cycling test and 4000-m cycling TT) on separate days following the completion of the long-graded exercise test. Subjects commenced a standardized 20-min warm-up before each performance trial comprising 5 min of cycling at 150 W, 8 min of cycling at a power output equivalent to 60% of the power output achieved at $\text{VO}_{2\text{max}}$, 2 min of cycling at a self-selected power output, 2 min of cycling to include three, 6-s maximal sprints and finished with 3 min of cycling at a self-selected power output. Subjects were then instructed to sit passively for a period of 8 min before starting the performance test. The average typical error of measurement for the performance variables from each performance test (expressed as a CV% between the familiarization and pre-supplementation performance tests) was mean power output 2.3% and performance time 1.2% (4000-m TT) and TTE 6.6% (supramaximal cycling test). A blood sample was taken from the earlobe pre-exercise (immediately before the performance trial), post-exercise and 5 min post-exercise for each supramaximal cycling test and 4000-m TT for determination of blood $[\text{La}^-]$ using the Lactate ProTM (Arkay KDK, Japan). An additional blood sample was collected into a preheparinized capillary tube at the same three time points on the day of each supramaximal cycling test and analysed for pH and $[\text{HCO}_3^-]$ using the i-STAT[®] blood gas analyser (i-STAT[®], Princeton, NJ). Gas-exchange was measured breath-by-breath using open-circuit spirometry (MedGraphics CPX/D, St. Paul, MN, USA) during the warm-up period and performance tests. The O_2 and CO_2 analyzers were calibrated before and after each test with precision reference gas, while calibration of the pneumotachograph was performed using a 3-L calibration syringe.

Supramaximal cycling tests

Before β -alanine supplementation, a supramaximal cycling test to exhaustion (PreExh) was conducted at the power output corresponding to 120% $\text{VO}_{2\text{max}}$ which was established by applying a linear regression to the steady-state VO_2 (min 4:00 - 5:00) and corresponding

power output data from the long-graded exercise test. Subjects commenced the standardized 20-min warm-up and then pedalled at the supramaximal power output ($\sim 110 \text{ rev} \cdot \text{min}^{-1}$) until they could no longer maintain a cadence $> 60 \text{ rev} \cdot \text{min}^{-1}$ despite strong verbal encouragement. Post-supplementation, the supramaximal cycling test to exhaustion (PostExh) was repeated using an identical protocol. In addition, subjects also completed another supramaximal cycling test (PostMatch) which was performed at the same power output, but the test was stopped at the time of exhaustion that was achieved during the pre-supplementation supramaximal cycling test. This provided a post-supplementation exhaustive (PostExh) and matched for work (PostMatch) comparison for PreExh. The order of PostMatch and PostExh was randomly assigned for each subject (Harmer et al. 2000).

4000-m cycling TT

A flat 4000-m TT profile was selected from the Velotron 3D Software and was displayed on a large monitor. The only instruction given to subjects was to complete the TT in the shortest amount of time possible. Subjects commenced the standardized warm-up before beginning each TT. Standardized verbal encouragement and feedback on the distance covered were given every 400 m. The gear ratio (53 x 17) was the same at the start of each TT, but subjects were permitted to adjust the gear ratio throughout the trial to reflect their preferred cadence.

Quantification of aerobic and anaerobic power

The aerobic and anaerobic contributions to power output were quantified from the total power output, VO_2 , respiratory-exchange ratio (RER) and gross efficiency by the methods used by Serresse et al. (1988; 1991) and adapted by Foster et al. (2003) which has recently been shown to be the most precise indirect estimate of the anaerobic contribution to total power output during a cycling TT (Noordhof et al. 2011). Power output and cardiorespiratory data were averaged and interpolated over 1-s intervals during the 4000-m TT and

supramaximal cycling tests. Split times were recorded for each 400-m interval completed during the 4000-m TT and for each 10-s interval during the supramaximal cycling tests. Subsequently, the average power output and the average VO_2 during each interval were calculated and the aerobic contribution to power output was calculated by multiplying metabolic work by gross efficiency. Gross efficiency was calculated from the average VO_2 and RER from the final 2 min of the warm-up stage that was completed at the power output equivalent to 60% of the power output achieved at $\text{VO}_{2\text{max}}$ (Garby and Astrup 1987). We assumed that $\text{RER} > 1.0$ were attributable to non-metabolic CO_2 production attributable to the buffering action by bicarbonate (Wasserman 1982). Accordingly, during the 4000-m TT and supramaximal cycling tests, RER values in excess of 1.0 were treated as if they equalled 1.0 in the calculation of metabolic work. The anaerobic contribution to power output was calculated by subtracting the power output attributable to aerobic metabolism from the total power output, for each interval. Summation of total power output, aerobic power, and anaerobic power over time provided measures of total work, aerobic work, and anaerobic work, respectively.

Training and dietary control

In an attempt to minimize any other potential diet-induced variability in exercise metabolism, subjects were instructed to consume the same types and quantities of food in the 24 h before each performance test. This was achieved by each participant recording a 24-h diet diary leading up to the first pre-supplementation performance trial, which was then replicated in the 24 h preceding each subsequent performance trial. Furthermore, each participant was asked to abstain from caffeine and alcohol and avoid strenuous exercise for the 24 h preceding each performance trial which was verbally confirmed with each subject prior to commencing each trial. Subjects were also asked to keep a record of all training sessions completed during the 28-d supplementation period, noting the activity, duration, and RPE. The RPE was measured

using the category ratio (CR10) Borg scale (Borg 1982) and multiplied by duration of each training session in minutes to provide a session-load RPE (Foster et al. 2001).

Statistical analysis

Statistical analyses were performed using IBM SPSS Statistics 19 (SPSS Inc, Chicago, USA). Data are presented as mean $\pm$ SD and the precision of the estimates of outcome statistics is shown as a 90% confidence limit unless otherwise stated. A fully-factorial ANOVA (group vs. trial) was conducted to investigate differences in the mean performance and physiological variables during the 4000-m TT and supramaximal cycling tests. To investigate the differences in pacing strategy during the 4000-m TT, the mean power output and the anaerobic and aerobic contribution to power output were calculated for each 400-m interval and statistically analysed using a three-way ANOVA (group v trial v distance covered). Where a significant interaction or main effect was observed, pairwise comparisons with Bonferroni adjustments were applied with significance being accepted at $P < 0.05$. These data were then further analysed using a modified statistical spreadsheet (Hopkins 2007), which calculates the likelihood (in percentage terms) of a condition having a positive/trivial/negative effect. The smallest worthwhile change was calculated using the between-athlete standard deviation (e.g., 0.2 multiplied by the between-athlete SD from the pre-supplementation tests) (Hopkins et al. 2009). Qualitative descriptors were assigned to the quantitative percentile scores as follows: 25-75% *possible*; 75-95% *likely*; 95-99% *very likely*; >99% *almost certain*. Where the chance of benefit or harm were both >5%, the true effect was deemed *unclear* (Batterham and Hopkins 2005).

Results:

Supramaximal cycling tests

There was no between group difference in TTE prior to supplementation (PreExh) in the supramaximal cycling test ($P = 0.449$; placebo: 176 ± 32 s and β -alanine: 174 ± 34 s) (Table 1). After supplementation (PostExh), there was a significant group $\times$ time interaction for TTE ($P = 0.041$), anaerobic capacity ($P = 0.048$) and blood $[\text{La}^-]$ ($P = 0.050$). Eight of the nine participants in the β -alanine group had a significant increase in TTE (change $\pm 90\%$ CL; $+12.6 \pm 8.2$ s; 90% likelihood; $P = 0.021$) with a corresponding increase in anaerobic capacity ($+1.1 \pm 0.7$ kJ; 85% likelihood; $P = 0.017$) (Figure 1) and post-exercise blood $[\text{La}^-]$ ($+1.1 \pm 0.9$ mM; 91% likelihood; $P = 0.041$), whilst there was no change in the placebo group. Seven of the nine subjects in the β -alanine group exceeded their pre-supplementation TTE by a greater magnitude than the smallest worthwhile change ($>6.9\%$), with one subject marginally improving ($+2.7\%$) and one subject performing worse (-5.9%). There was no difference in either group for $\text{VO}_{2\text{peak}}$ ($\text{L} \cdot \text{min}^{-1}$), or gross efficiency measured during the preceding warm-up period prior ($P > 0.05$).

The matched-work comparison (PostMatch) following the supplementation period revealed no between- or within-group differences in anaerobic capacity, blood $[\text{La}^-]$, $\text{VO}_{2\text{peak}}$ or gross efficiency measured during the preceding warmup between groups ($P > 0.05$). There was no significant interaction effect for the absolute pH values at any time point but the fall in blood pH from pre- to post-exercise in PostMatch tended to be less pronounced in the β -alanine group compared to the placebo group (a difference of -0.029 ± 0.035 units between the $\Delta \beta$ -alanine - Δ Placebo; $P = 0.080$).

There were no significant alterations to pre-exercise blood pH, $[\text{La}^-]$ or $[\text{HCO}_3^-]$ following the supplementation period in either the placebo or β -alanine group (Table 2). In all supramaximal cycling tests, blood pH and $[\text{HCO}_3^-]$ were significantly reduced and blood $[\text{La}^-]$ was significantly elevated from pre- to post-exercise and after 5 min of recovery in both groups ($P < 0.001$).

4000-m TT

In the pre-supplementation 4000-m TT there was no difference in performance time ($P = 0.457$; placebo: 360.0 ± 16.3 s and β -alanine: 361.5 ± 20.5 s) or mean power output ($P = 0.456$; placebo: 335.8 ± 28.1 W and β -alanine: 332.8 ± 26.1 W) between the placebo and β -alanine group (Table 3). After supplementation, there was a significant group $\times$ time interaction for performance time ($P = 0.034$) and mean power output ($P = 0.043$) with the β -alanine group reducing performance time (-5.9 ± 5.7 s; 94% likelihood; $P = 0.014$) and increasing mean power output ($+11.1 \pm 8.7$ W; 89% likelihood; $P = 0.025$), whilst there was no change in the placebo group (Figure 2).

Analysis of the differences in the distribution of the aerobic and anaerobic contributions to energy provision revealed a significant increase in the whole trial mean anaerobic power output for the β -alanine group (a difference of $+6.2 \pm 4.5$ W between the $\Delta \beta$ -alanine - Δ Placebo; 89% likelihood; $P = 0.035$) (Table 3), whilst there was no change in the placebo group. Pairwise comparisons revealed that the anaerobic contribution to power output was significantly greater at the 2000- and 2400-m intervals in the β -alanine group (Figure 3). The aerobic contribution to power output was not significantly different for either group at any time point ($P > 0.05$) (Figure 4). There was no difference for either group for $\text{VO}_{2\text{peak}}$, blood $[\text{La}^-]$ or RPE ($P > 0.05$).

Discussion:

The present study employed a rigorous methodological approach to investigate the effects of β -alanine supplementation on the resultant blood acidosis, lactate accumulation and energy provision during work-matched and exhaustive supramaximal-intensity cycling, as well as the aerobic and anaerobic contribution to power output during a 4000-m cycling TT. We demonstrated that β -alanine supplementation had no effect on anaerobic capacity during a

work-matched bout of supramaximal cycling. We also showed that TTE during supramaximal cycling to exhaustion was improved, concomitant with increased blood $[La^-]$ and anaerobic capacity, suggesting that the volume of anaerobic attributable energy is enhanced by β -alanine supplementation. Furthermore, the time required to complete a 4000-m cycling TT was reduced, accompanied by a higher anaerobic contribution to power output during in the middle of the TT.

In the present study, our matched work supramaximal cycling test comparison allowed for analysis of changes in energy provision, blood $[La^-]$ and blood pH under conditions of identical work before and after β -alanine supplementation. In comparison with the placebo group, β -alanine supplementation resulted in a trend towards an attenuation of the exercise-induced blood acidosis which was accompanied by an unchanged blood $[La^-]$ and anaerobic capacity. This is in agreement with the data from Baguet et al. (2010b) who reported a reduction in exercise-induced blood acidosis following a 6-min exercise bout at a fixed intensity (50% of the difference between ventilatory threshold and VO_{2peak}) after supplementation with β -alanine, without affecting blood $[La^-]$ and $[HCO_3^-]$ concentrations. In opposition to the findings of the present study, and that of Baguet et al. (2010b), Gross et al. (2014) reported a reduced accumulated oxygen deficit ($-5.0 \pm 5.0\%$, $d = 0.6$), concurrent with an increased aerobic energy contribution ($1.4 \pm 1.3\%$, $d = 0.5$) during a 90-s bout of cycling (110% peak power) following β -alanine supplementation. The mechanism that may support an increased contribution of aerobic energy production during fixed duration/fixed power output cycling tests following β -alanine supplementation is faster oxygen uptake kinetics or an increased peak oxygen uptake (Whipp et al. 1982). However, Gross et al (2014) reported no change in peak oxygen uptake following β -alanine supplementation and Baguet et al. (2010b) reported no change in the overall fast component of VO_2 kinetics following supplementation with β -alanine. A factor that may explain the discrepancy between the

findings of the present study, and that of Gross et al. (2014), is the method used to quantify anaerobic capacity. The present study employed the gross efficiency method (Serresse et al. 1988; Serresse et al. 1991; Foster et al. 2003), whilst Gross et al. (2014) preferred the accumulated oxygen deficit method (Medbo et al. 1988) to quantify anaerobic capacity. The accumulated oxygen deficit method of quantifying anaerobic attributable energy has been criticized for having limitations as a valid and reliable measure of anaerobic capacity (Noordhof et al. 2010). Furthermore, recent research (Noordhof et al. 2010; Noordhof et al. 2011) has suggested the use of 10×4 min submaximal exercise bouts and a fixed value of the y-intercept would result in the most robust power output- VO_2 relationship and should therefore result in the most valid and reliable results for the quantification of anaerobic capacity using the accumulated oxygen deficit method. The study of Gross et al. (2014) employed only 2 min stages during a graded exercise test to exhaustion to develop the power output- VO_2 relationship from which the VO_2 demand of the supramaximal exercise bout was predicted. These methodological shortcomings with regards to the calculation of the accumulated oxygen deficit may introduce meaningful errors in the quantification of anaerobic capacity.

The longer TTE following β -alanine supplementation observed in the present study was accompanied by a significantly larger anaerobic capacity and a significantly higher blood $[\text{La}^-]$. This demonstrates that anaerobic capacity measured over the entire exercise bout may be enhanced following β -alanine supplementation, concomitant with improved performance during supramaximal, exhaustive exercise. Given that the values for anaerobic capacity were similar when the work was matched, it could be suggested that the greater anaerobic capacity during exhaustive supramaximal exercise following supplementation was a consequence of the prolonged exercise duration and not a faster rate of anaerobic energy production. The beneficial effect of supplementation with β -alanine on muscular performance is thought to

stem from carnosine's undisputable role as an intracellular buffer (Bate-Smith 1938), effectively attenuating the rate at which intracellular pH falls during exercise (Baguet et al. 2010b). If augmentation of muscle carnosine is commensurate with an enhanced buffer capacity, the attainment of a critical state of muscle acidosis could be attenuated, enabling muscle contractions and exercise to be performed for a longer duration before the onset of fatigue. However, it should also be noted that changes in buffer capacity may not be the sole mechanism by which any putative performance-enhancing effects of augmented carnosine are manifested. Indeed, β -alanine supplementation may enhance muscle contractile properties by possibly aiding Ca^{2+} release from type I muscle fibres (Dutka et al. 2012). However, a recent *in vivo* study with humans did not support this mechanism, but suggested that augmentation of carnosine resulted in an improvement in calcium reuptake (Hannah et al. 2015). Further research is required to elucidate the mechanism(s) that may support the ergogenic effects of supplementation with β -alanine.

The current study demonstrated a significant improvement in 4000-m TT time by an average of $1.7 \pm 1.3\%$ (-6.3 ± 4.6 s) which is supported by previous research investigating the effects of β -alanine supplementation on TT that are 1-7 min in duration (De Salles Painelli et al. 2013; Ducker et al. 2013; Hobson et al. 2013; Bellinger and Minahan 2015). β -alanine supplementation has recently been shown to be *very likely* beneficial to 2000-m rowing performance (412.1 ± 13.2 s) (Hobson et al. 2013) and significantly improve 200-m swimming performance (131.6 ± 8.8 s) (De Salles Painelli et al. 2013) and 800-m track running performance (145.7 ± 5.7 s) (Ducker et al. 2013). The distribution of the anaerobic power output during short-duration TT (1-7 min) has been considered the main metabolic pathway determining both pacing strategy (Hettinga et al. 2006; Hettinga et al. 2007) and performance (Aisbett et al. 2009) during such events. In the present study, the augmented anaerobic capacity observed during the exhaustive supramaximal cycling test following β -

alanine supplementation was mirrored by an increase in the mean anaerobic power output during the 4000-m TT with the most pronounced increases in anaerobic power output occurring at the 2000- and 2400-m interval. Given that the amount of anaerobic energy that can be produced during a TT is a constant volume (i.e. the anaerobic capacity) (Hettinga et al. 2006; Hettinga et al. 2007), it could be expected that the anaerobic power output at the beginning or the end of the TT would be reduced in the β -alanine group to compensate for the greater anaerobic power output in the middle of the TT. Instead, the β -alanine group were able to produce a similar fast start, followed by a transition to an even pacing strategy, with an increased anaerobic power output, and finished the TT with a similar end spurt. It could be hypothesized that the H^+ accumulation during the intense fast start may have been blunted by the additional buffering effects of β -alanine supplementation (via carnosine) which had the greatest influence during this period. The enhanced pH homeostasis may have allowed for an increase in the anaerobic power output, plausibly via an effect associated with increased muscle glycogenolysis and glycolysis, resulting in an enhanced overall performance. This supposition is supported by Baguet et al (2010a), who demonstrated that higher intramuscular carnosine concentrations were correlated with faster rowing times over the second and third 500-m splits (~ 1.5 – 5 min) of a 2000-m rowing TT and hypothesized that this could be due to enhanced muscle buffering capacity over this intense period of the TT.

A limitation of the current study is that we did not measure muscle carnosine content which would have quantified the efficacy of our supplementation protocol ($6.4 \text{ g} \cdot \text{day}^{-1}$ for 28 d). However, our β -alanine supplementation protocol is similar to that used by Hill et al. (2005), whose participants consumed $4.0 - 6.4 \text{ g} \cdot \text{day}^{-1}$ of β -alanine for 28 d and exhibited a significant increase in muscle carnosine content of 59% (vastus lateralis). Furthermore, Bex et al., (2014) demonstrated that carnosine loading in response to supplementation with β -alanine is more pronounced in the trained vs. untrained muscles of athletes by reporting a

significantly greater increase in muscle carnosine in the leg muscles (soleus and gastrocnemius) of cyclists ($+0.084 \pm 0.027$ AU) compared to nonathletes ($+0.051 \pm 0.031$ AU). From this data we can be confident in presuming that the supplementation protocol employed in the current study would significantly elevate levels of muscle carnosine content.

In summary, we present findings demonstrating that β -alanine supplementation significantly extended TTE during exhaustive supramaximal cycling, concomitant with an augmented anaerobic capacity and blood $[La^-]$. Considering that β -alanine supplementation did not alter anaerobic capacity during a work-matched cycling test, the increased anaerobic capacity during exhaustive supramaximal cycling was a consequence of the prolonged TTE. We also showed that cyclists were able to perform a 4000-m TT faster following supplementation with β -alanine, commensurate with a greater anaerobic contribution to power output during the middle of the TT.

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References:

Abbiss, C.R., Levin, G., McGuigan, M.R., and Laursen, P.B. 2008. Reliability of power output during dynamic cycling. *Int. J. Sports Med.* **29**: 574-578.

Abe, H. 2000. Role of histidine-related compounds as intracellular proton buffering constituents in vertebrate muscle. *Biochemistry (Mosc.)* **65**: 757-765.

Aisbett, B., Le Rossignol, P., McConell, G.K., Abbiss, C.R., and Snow, R. 2009. Effects of starting strategy on 5-min cycling time-trial performance. *J. Sports Sci.* **27**: 1201-1209.

- Baguet, A., Bourgois, J., Vanhee, L., Achten, E., and Derave, W. 2010a. Important role of muscle carnosine in rowing performance. *J. Appl. Physiol.* **109**: 1096-1101.
- Baguet, A., Koppo, K., Pottier, A., and Derave, W. 2010b. β -Alanine supplementation reduces acidosis but not oxygen uptake response during high-intensity cycling exercise. *Eur. J. Appl. Physiol.* **108**: 495-503.
- Bate-Smith, E. 1938. The buffering of muscle in rigour: protein, phosphate and carnosine. *J. Physiol.* **92**: 336-343.
- Batterham, A.M. and Hopkins, W.G. 2005. Making meaningful inferences about magnitudes. *Sportscience* **9**: 6-13.
- Bellinger, P.M. 2014. β -alanine supplementation for athletic performance: An update. *J. Strength Cond. Res.* **28**: 1751–1770.
- Bellinger, P.M. and Minahan, C.L. 2015. The effect of β -alanine supplementation on cycling time trials of different length. *Eur. J. Sport Sci.* (Epub ahead of print) DOI:10.1080/17461391.2015.1120782
- Bex, T., Chung, W., Baguet, A., Stegen, S., Stautemas, J., Achten, E., and Derave, W. 2014. Muscle carnosine loading by β -alanine supplementation is more pronounced in trained vs. untrained muscles. *J. Appl. Physiol.* **116**: 204-209.
- Blancquaert, L., Everaert, I., and Derave, W. 2015. β -alanine supplementation, muscle carnosine and exercise performance. *Curr. Opin. Clin. Nutr. Metab. Care* **18**: 63-70.
- Borg, G.A.v. 1982. Psychophysical bases of perceived exertion. *Med. Sci. Sports Exerc.* **14**: 377-381.

Craig, N.P., Norton, K.I., Bourdon, P.C., Woolford, S., Stanef, T., Squires, B., Olds, T.S., Conyers, R., and Walsh, C. 1993. Aerobic and anaerobic indices contributing to track endurance cycling performance. *Eur. J. Appl. Physiol.* **67**: 150-158.

De Salles Painelli, V., Roschel, H., de Jesus, F., Sale, C., Harris, R.C., Solis, M.Y., Benatti, F.B., Gualano, B., Lancha, A.H., and Artioli, G.G. 2013. The ergogenic effect of β -alanine combined with sodium bicarbonate on high-intensity swimming performance. *Appl. Physiol. Nutr. Metab.* **38**: 525-532.

Donovan, T., Ballam, T., Morton, J.P., and Close, G.L. 2012. β -alanine improves punch force and frequency in amateur boxers during a simulated contest. *Int. J. Sport Nutr. Exerc. Metab.* **22**: 331-337.

Ducker, K.J., Dawson, B., and Wallman, K.E. 2013. Effect of β -alanine supplementation on 800-m running performance. *Int. J. Sport Nutr. Exerc. Metab.* **23**: 554-561.

Dutka, T.L., Lambole, C.R., McKenna, M.J., Murphy, R.M., and Lamb, G.D. 2012. Effects of carnosine on contractile apparatus Ca^{2+} sensitivity and sarcoplasmic reticulum Ca^{2+} release in human skeletal muscle fibers. *J. Appl. Physiol.* **112**: 728-736.

Foster, C., de Koning, J.J., Hettinga, F., Lampen, J., La Clair, K.L., Dodge, C., Bobbert, M., and Porcari, J.P. 2003. Pattern of energy expenditure during simulated competition. *Med. Sci. Sports Exerc.* **35**: 826-831.

Foster, C., Florhaug, J.A., Franklin, J., Gottschall, L., Hrovatin, L.A., Parker, S., Doleshal, P., and Dodge, C. 2001. A new approach to monitoring exercise training. *J. Strength Cond. Res.* **15**: 109-115.

- Garby, L. and Astrup, A. 1987. The relationship between the respiratory quotient and the energy equivalent of oxygen during simultaneous glucose and lipid oxidation and lipogenesis. *Acta Physiol. Scand.* **129**: 443-444.
- Gross, M., Boesch, C., Bolliger, C., Norman, B., Gustafsson, T., Hoppeler, H., and Vogt, M. 2014. Effects of β -alanine supplementation and interval training on physiological determinants of severe exercise performance. *Eur. J. Appl. Physiol.* **114**: 221-234.
- Hannah, R., Stannard, R.L., Minshull, C., Artioli, G.G., Harris, R.C., and Sale, C. 2015. β -alanine supplementation enhances human skeletal muscle relaxation speed but not force production capacity. *J. Appl. Physiol.* **118**: 604-612.
- Harmer, A.R., McKenna, M.J., Sutton, J.R., Snow, R.J., Ruell, P.A., Booth, J., Thompson, M.W., Mackay, N.A., Stathis, C.G., Crameri, R.M., Carey, M.F., and Eager, D.M. 2000. Skeletal muscle metabolic and ionic adaptations during intense exercise following sprint training in humans. *J. Appl. Physiol.* **89**: 1793-1803.
- Hettinga, F.J., De Koning, J.J., Broersen, F.T., Van Geffen, P., and Foster, C. 2006. Pacing strategy and the occurrence of fatigue in 4000-m cycling time trials. *Med. Sci. Sports Exerc.* **38**: 1484-1491.
- Hettinga, F.J., De Koning, J.J., Meijer, E., Teunissen, L., and Foster, C. 2007. Effect of pacing strategy on energy expenditure during a 1500-m cycling time trial. *Med. Sci. Sports Exerc.* **39**: 2212-2218.
- Hill, C., Harris, R., Kim, H., Boobis, L., Sale, C., and Wise, J. 2005. The effect of beta-alanine and creatine monohydrate supplementation on muscle composition and exercise performance. *Med. Sci. Sports Exerc.* **37**: S348.

- Hill, C.A., Harris, R.C., Kim, H.J., Harris, B.D., Sale, C., Boobis, L.H., Kim, C.K., and Wise, J.A. 2007. Influence of β -alanine supplementation on skeletal muscle carnosine concentrations and high intensity cycling capacity. *Amino Acids* **32**: 225-233.
- Hobson, R.M., Harris, R.C., Martin, D., Smith, P., Macklin, B., Gualano, B., and Sale, C. 2013. Effect of β -alanine, with and without sodium bicarbonate, on 2000-m rowing performance. *Int. J. Sport. Nutr. Exerc. Metab.* **23**: 480-487.
- Hopkins, W. 2007. A spreadsheet for deriving a confidence interval, mechanistic inference and clinical inference from a P value. *Sportscience* **11**: 16-20.
- Hopkins, W., Marshall, S., Batterham, A., and Hanin, J. 2009. Progressive statistics for studies in sports medicine and exercise science. *Med. Sci. Sports Exerc.* **41**: 3-12.
- Medbo, J.I., Mohn, A.C., Tabata, I., Bahr, R., Vaage, O., and Sejersted, O.M. 1988. Anaerobic capacity determined by maximal accumulated O₂ deficit. *J. Appl. Physiol.* **64**: 50-60.
- Noordhof, D., de Koning, J., and Foster, C. 2010. The maximal accumulated oxygen deficit method: A valid and reliable measure of anaerobic capacity? *Sports Med.* **40**: 285-302.
- Noordhof, D.A., Vink, A.M.T., de Koning, J.J., and Foster, C. 2011. Anaerobic capacity: effect of computational method. *Int. J. Sports Med.* **32**: 422-428.
- Ramsbottom, R., Nevill, A.M., Nevill, M.E., Newport, S., and Williams, C. 1994. Accumulated oxygen deficit and short-distance running performance. *J. Sports Sci.* **12**: 447-453.
- Sale, C., Artioli, G., Gualano, B., Saunders, B., Hobson, R., and Harris, R. 2013. Carnosine: from exercise performance to health. *Amino Acids* **44**: 1477-1491.

Sale, C., Saunders, B., Hudson, S., Wise, J.A., Harris, R.C., and Sunderland, C.D. 2011. Effect of β -alanine plus sodium bicarbonate on high-intensity cycling capacity. *Med. Sci. Sports Exerc.* **43**: 1972-1978.

Serresse, O., Lortie, G., Bouchard, C., and Boulay, M.R. 1988. Estimation of the contribution of the various energy systems during maximal work of short duration. *Int. J. Sports Med.* **9**: 456-460.

Serresse, O., Simoneau, J.A., Bouchard, C., and Boulay, M.R. 1991. Aerobic and anaerobic energy contribution during maximal work output in 90 s determined with various ergocycle workloads. *Int. J. Sports Med.* **12**: 543-547.

Sporer, B. and McKenzie, D. 2007. Reproducibility of a laboratory based 20-km time trial evaluation in competitive cyclists using the Velotron Pro ergometer. *Int. J. Sports Med.* **28**: 940-944.

Stellingwerff, T., Decombaz, J., Harris, R., and Boesch, C. 2012. Optimizing human in vivo dosing and delivery of β -alanine supplements for muscle carnosine synthesis. *Amino Acids* **43**: 57-65.

Tobias, G., Benatti, F., Salles Painelli, V., Roschel, H., Gualano, B., Sale, C., Harris, R., Lancha, A., Jr., and Artioli, G. 2013. Additive effects of β -alanine and sodium bicarbonate on upper-body intermittent performance. *Amino Acids* **45**: 309-317.

Wasserman, K. 1982. Dyspnea on exertion: Is it the heart or the lungs? *JAMA* **248**: 2039-2043.

Weber, C.L. and Schneider, D.A. 2002. Increases in maximal accumulated oxygen deficit after high-intensity interval training are not gender dependent. *J. Appl. Physiol.* **92**: 1795-1801.

Whipp, B.J., Ward, S.A., Lamarra, N., Davis, J.A., and Wasserman, K. 1982. Parameters of ventilatory and gas exchange dynamics during exercise. *J. Appl. Physiol.* **52**: 1506-1513.

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Tables:

Table 1 - Mean and SEM values for $\text{VO}_{2\text{peak}}$, absolute and relative anaerobic capacity, gross efficiency and TTE measured during the supramaximal cycling tests to exhaustion conducted pre-supplementation (PreExh) and post-supplementation (PostExh) as well as during the additional post-supplementation supramaximal cycling test identical in duration and power output to PreExh (PostMatch) for the β -alanine and placebo group.

		PreExh	PostMatch	PostExh
TTE (s)	β -alanine	174 ± 34	173 ± 34	$187 \pm 35^{*\dagger}$
	Placebo	176 ± 32	175 ± 33	176 ± 34
Anaerobic capacity (kJ)	β -alanine	19.8 ± 3.4	19.9 ± 3.4	$21.2 \pm 3.6^{*\dagger}$
	Placebo	19.8 ± 3.9	19.6 ± 3.8	19.9 ± 3.7
Anaerobic capacity ($\text{J} \cdot \text{kg}^{-1}$)	β -alanine	284 ± 61	284 ± 61	$298 \pm 61^{*\dagger}$
	Placebo	286 ± 62	283 ± 61	287 ± 60
VO_2 peak ($\text{L} \cdot \text{min}^{-1}$)	β -alanine	4.26 ± 0.50	4.24 ± 0.60	4.27 ± 0.61
	Placebo	4.29 ± 0.59	4.30 ± 0.59	4.32 ± 0.56
Gross efficiency (%)	β -alanine	21.6 ± 1.1	21.4 ± 1.4	21.6 ± 1.7
	Placebo	21.5 ± 1.1	21.5 ± 1.2	21.4 ± 1.5

The data for gross efficiency was measured during the final 2 minutes of the warm up stage that was completed at 60% of peak aerobic power output that preceded each supramaximal cycling bout.

*Denotes a significant difference from PostMatch ($P < 0.05$).

† Denotes a significant difference from PreExh ($P < 0.05$).

TTE = time to exhaustion.

Table 2 - Mean and SEM values for pH, blood [La⁻] and [HCO₃⁻] concentrations measured at pre-exercise (immediately before the performance trial), post-exercise and 5 min post-exercise for each supramaximal cycling bout.

	Placebo			β-alanine		
	PreExh	PostMatch	PostExh	PreExh	PostMatch	PostExh
pH						
Pre-exercise	7.437 ± 0.094	7.427 ± 0.071	7.433 ± 0.069	7.433 ± 0.041	7.431 ± 0.037	7.436 ± 0.026
Post-exercise	7.097 ± 0.035*	7.091 ± 0.063*	7.092 ± 0.055*	7.096 ± 0.056*	7.107 ± 0.053*	7.101 ± 0.065*
Post-exercise (+5 min)	7.102 ± 0.037*	7.098 ± 0.065*	7.099 ± 0.069*	7.100 ± 0.053*	7.115 ± 0.045*	7.099 ± 0.060*
ΔPre to post-exercise	-0.331 ± 0.056	-0.347 ± 0.064	-0.342 ± 0.052	-0.337 ± 0.065	-0.324 ± 0.072	-0.335 ± 0.079
Lactate (mmol·L ⁻¹)						
Pre-exercise	1.8 ± 0.4	2.0 ± 0.4	1.9 ± 0.5	1.9 ± 0.4	1.8 ± 0.4	2.0 ± 0.5
Post-exercise	14.9 ± 3.0*	14.9 ± 2.6*	14.8 ± 2.7*	14.6 ± 2.3*	14.3 ± 2.9*	15.6 ± 2.5*
Post-exercise (+5 min)	13.6 ± 2.7*	13.5 ± 2.6*	13.9 ± 2.3*	13.3 ± 1.9*	13.5 ± 3.1*	13.8 ± 1.9*
ΔPre to post-exercise	13.0 ± 2.7	12.9 ± 2.8	12.9 ± 2.9	12.7 ± 2.5	12.5 ± 3.1	13.6 ± 2.0†\$
HCO ₃ ⁻ (mmol·L ⁻¹)						
Pre-exercise	26.59 ± 1.29	26.93 ± 1.71	27.18 ± 2.33	26.99 ± 1.62	26.32 ± 1.69	26.54 ± 1.14
Post-exercise	12.16 ± 2.43*	12.01 ± 2.53*	11.10 ± 1.80*	10.66 ± 1.98*	10.86 ± 1.69*	10.89 ± 1.40*
Post-exercise (+5 min)	10.54 ± 2.12*	10.76 ± 1.97*	10.22 ± 1.97*	9.84 ± 1.93*	9.97 ± 2.14*	10.03 ± 1.97*
ΔPre to post-exercise	-14.95 ± 2.91	-15.10 ± 2.59	-14.72 ± 2.09	-14.43 ± 3.64	-14.92 ± 3.49	-15.08 ± 2.28

* Denotes a significant effect of time from pre-exercise (P < 0.0001).

† Denotes a significant effect of trial from PreExh (P < 0.05).

\$ Denotes a significant effect of trial from PostMatch (P < 0.05).

Table 3 - Physiological and performance variables during the 4000-m cycling TT for the β -alanine and placebo group.

Variable	Placebo		β -alanine		ANOVA Group X Trial interaction, P value	$\Delta \beta$ -alanine - Δ Placebo raw difference (90%CL)	Likelihood of β -alanine being positive/trivial/negative (compared with Placebo)
	Pre	Post	Pre	Post			
Performance time (s)	360.0 $\pm$ 16.3	360.4 $\pm$ 14.8	361.5 $\pm$ 20.5	355.6 $\pm$ 22.4 [†]	0.034	-6.3 $\pm$ 4.6*	85 / 15 / 0
Mean power output (W)	335.8 $\pm$ 28.1	334.5 $\pm$ 29.2	332.8 $\pm$ 26.1	342.6 $\pm$ 27.9 [†]	0.043	11.0 $\pm$ 8.7*	89 / 11 / 0
Anaerobic power output (W)	62.4 $\pm$ 16.7	62.0 $\pm$ 19.9	61.6 $\pm$ 15.8	67.4 $\pm$ 17.9 [†]	0.035	6.2 $\pm$ 4.5*	89 / 11 / 0
Aerobic power output (W)	273.4 $\pm$ 16.0	272.5 $\pm$ 15.8	271.2 $\pm$ 14.8	273.6 $\pm$ 13.0	0.328	4.8 $\pm$ 8.6	29 / 70 / 1
VO _{2peak} (L·min ⁻¹)	4.43 $\pm$ 0.53	4.44 $\pm$ 0.65	4.42 $\pm$ 0.61	4.45 $\pm$ 0.60	0.823	0.02 $\pm$ 0.16	17 / 75 / 8
Blood [La-] (mmol·L ⁻¹)	15.3 $\pm$ 3.3	15.2 $\pm$ 3.0	14.4 $\pm$ 2.6	15.1 $\pm$ 2.4	0.326	0.40 $\pm$ 0.72	60 / 45 / 5
RPE (AU)	18.5 $\pm$ 0.9	18.8 $\pm$ 0.7	18.7 $\pm$ 1.2	18.8 $\pm$ 1.1	0.736	0.10 $\pm$ 0.54	16 / 88 / 6

*Denotes a significant group X trial interaction ($P < 0.05$).

[†] Denotes a significant difference from pre-supplementation ($P < 0.05$).

Figure captions:

Figure 1 - Mean and SEM values for anaerobic capacity for the β -alanine (panel A) and placebo (panel B) group measured during the supramaximal cycling tests to exhaustion conducted pre-supplementation (PreExh) and post-supplementation (PostExh) as well as during the additional post-supplementation supramaximal cycling test identical in duration and power output to PreExh (PostMatch). * Denotes a significant difference from PostMatch ($P < 0.05$). † Denotes a significant difference from PreExh ($P < 0.05$).

Figure 2 - Mean and SEM values for the mean power output profile during the 4000-m TT for each 400-m interval for the β -alanine (panel A) and placebo group (panel B). * Denotes a significant difference from pre-supplementation ($P < 0.05$).

Figure 3 - Mean and SEM values for the anaerobic contribution to the total power output profile during the 4000-m TT for each 400-m interval for the β -alanine (panel A) and placebo group (panel B). * Denotes a significant difference from pre-supplementation ($P < 0.05$).

Figure 4 - Mean and SEM values for the aerobic contribution to the total power output profile during the 4000-m TT for each 400-m interval for the β -alanine (panel A) and placebo group (panel B).

Figure 1 - Mean and SEM values for anaerobic capacity for the β -alanine (panel A) and placebo (panel B) group measured during the supramaximal cycling tests to exhaustion conducted pre-supplementation (PreExh) and post-supplementation (PostExh) as well as during the additional post-supplementation supramaximal cycling test identical in duration and power output to PreExh (PostMatch). * Denotes a significant difference from PostMatch ($P < 0.05$). † Denotes a significant difference from PreExh ($P < 0.05$).
297x209mm (300 x 300 DPI)

Figure 2 - Mean and SEM values for the mean power output profile during the 4000-m TT for each 400-m interval for the β -alanine (panel A) and placebo group (panel B). * Denotes a significant difference from pre-supplementation (P < 0.05).
239x89mm (300 x 300 DPI)

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Metabolic consequences of β -alanine supplementation during exhaustive supramaximal cycling and 4000-m time trial performance

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