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Influence of Absolute versus relative L-arginine Dosage on 1 km and 16.1 km time trial performance in trained cyclists

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Abstract

This investigation aimed to determine the effects of L-arginine supplementation on cycling time trial (TT) performance. Eight trained male cyclists performed 1 and 16.1km time trials on three occasions, control (CON), absolute (ABS) and relative (REL) loading. Participants consumed 500ml of water with either 6g (ABS), 0.15 g·kg^{-1} body mass (REL) of L-arginine or water (CON) 90min prior to testing. Time to completion, mean power output (W_{mean}) and post-exercise lactate (La) were recorded for each TT. Time to completion decreased non-significantly for 1 and 16.1km TT's during ABS and REL trials compared to CON. W_{mean} was significantly different between CON and REL during 16.1 km TT (196.19 ± 32.40W and 215.81 ± 31.56W). Blood lactates was significantly different between CON and ABS for the 1 km TT (p = 0.04) (13.59 ± 1.21 mmol·L^{-1} and 12.38 ± 0.70 mmol·L^{-1}, respectively) and between CON and ABS (p = 0.04) (9.11 ± 2.91mmol·L^{-1} and 7.64 ± 3.01mmol·L^{-1}, respectively) and CON and REL (9.11 ± 2.91mmol·L^{-1} and 7.15 ± 2.96mmol·L^{-1}, respectively) for 16.1km TT. These results indicate L-arginine supplementation does not significantly improve cycling TT performance, though there was a trend towards reduced time to completion and increased mean power output, and that relative doses appear more effective than absolute doses.

Keywords: physiology, performance, nutrition, supplementation.

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Introduction

L-arginine (ARG) is a conditionally essential amino acid and has numerous metabolic roles and vascular control (McConell, 2007; Morris, 2007; Wu et al. 2009). One such role, as a pre-cursor to synthesise nitric oxide (NO) via the L-arginine-nitric oxide pathway (Moncada and Higgs, 1993), has been proposed to improve exercise performance through enhanced neurotransmission and vasodilation (Böger and Bode-Böger, 2001).

Several studies have investigated the effects of ARG on exercise parameters. Improvements in exercise parameters have been observed when supplementing with ARG alone (Schaefer et al. 2002) or when mixed with other compounds including other amino acids and vitamins (Bailey et al. 2010a), α-ketoglutarate (Campbell et al. 2006), hydrochloride (Koppo et al. 2009) and grape seed extract (Camic et al. 2010). Moreover, research has observed reductions in blood lactate (La) (Schaefer et al. 2002), increases in peak power output (Campbell et al. 2006) and time to exhaustion (Bailey et al. 2010a). Álvares et al. (2012) suggested that ARG supplementation increases the availability of ARG within the system, thereby enhancing synthesis of NO via the enzyme endothelial nitric oxide synthase (eNOS). However, research supporting this has only elicited increases in NO in diseased populations (Schellong et al. 1997), or when supplementing ARG with other compounds (Bailey et al. 2010a).

However, the relationship between eNOS and ARG is somewhat controversial and the availability of ARG appears to be a rate-limiting factor in NO synthesis (Flam et al. 2007). Whilst circulating ARG concentrations are typically between 50-200 µM (Wu and Morris, 1998) intracellular concentrations are as high as 800 µM (Baydoun et al. 1990). However, the half-saturating concentration (K_M) for ARG has been reported to be ~2.9 µM, indicating that eNOS operates under saturated concentrations and that the majority of intracellular ARG may not be available for conversion to NO (Hardy and May, 2002). Despite this, several studies have shown supplementation with ARG to augment endothelial NO production (Hishikawa et al. 1993; Böger et al. 1994; Alvares et al. 2012) . As such this relationship has been termed the "arginine-paradox".

An increase in vasodilation and blood flow as a result of ARG supplementation would potentially increase both nutrient delivery (Schellong et al. 1997) and lactate clearance by the muscles (Schaefer et al. 2002), thus improving tolerance to physical activity and aiding recovery (Bescós et al. 2012). In addition, McConell and Kingwell (2006) found that infusion of ARG resulted in enhanced glucose uptake by skeletal muscle, hypothesising that this was due to increased eNOS activity and production of NO, thereby increasing

glucose disposal and oxidative phosphorylation during exercise and inhibiting glycolysis. However, this process is far more complex, as Benavides et al. (2013) found low fluxes of NO stimulated glycolysis and in fact inhibited oxidative phosphorylation and that glycolytic pathways were highly dependent on oxygen tension. Therefore, the role of ARG and subsequently NO in metabolism is somewhat ambiguous and could exert either positive or negative influences on performance dependent upon the internal environment.

Research regarding metabolite removal is equivocal (Schaefer et al. 2002; Burtscher, Brunner, Faulhaber, Hotter and Likar, 2005; Liu et al. 2009; Olek et al. 2010). Schaefer et al. (2002) demonstrated metabolic changes after a 3 g ARG load with no additional compounds. However, ARG was administered intravenously, which would be impractical in a sport specific environment. Conversely, Liu et al. (2009) observed no significant differences in metabolic changes after orally supplementing 10 elite male judo athletes with 6 g ARG for 3 days. However, as the athletes in their study were all at elite level, the efficiency of metabolite removal may already have been at its optimum.

Bailey et al. (2010a) provide the only research to date that has investigated the effect of ARG supplementation on endurance performance. They demonstrated that an acute 6 g dose of ARG an hour prior to a series of moderate and severe-intensity exercise bouts for 3 days resulted in significant increases in plasma nitrite (NO_2^-) and time to failure. There was also a significant reduction in oxygen uptake (VO_2) during moderate intensity exercise, and VO_2 slow component amplitude during severe-intensity exercise following supplementation. However, the supplement administered contained trace amounts of vitamins E, C, B6, and B12 and other amino acids including L-glutamine, L-leucine, L-valine, L-carnitine, L-citrulline, L-cysteine, and L-isoleucine along with 11 g of fructose, all of which may have acted as ergogenic aids themselves

There is a lack of research to date that has observed significant differences during predominately anaerobic exercise after supplementation of ARG. However, Campbell et al. (2006) provides an insight into the possible benefits to anaerobic performance after supplementing 12 $g.day^{-1}$ of ARG α-ketoglutarate (AAKG) for 4 weeks. Results revealed significantly greater gains in strength during one repetition maximum bench press and peak power output during a 30 second Wingate test when compared to a control group. It was suggested that the improvements were a result of ARG also being a pre-cursor of creatine synthesis (Persky and Brazeau, 2001). However, the influence of α-ketoglutarate on the improvements observed cannot be discounted. All of the current literature that has elicited significant differences when examining ARG supplementation on exercise parameters have used ARG in combination with other compounds (Bailey et al. 2010a; Camic et al. 2010; Koppo et al. 2009; Campbell et al. 2006).

Body size is known to influence physiological responses to ingested supplements (Jeacocke and Burke, 2010). It is possible that a relative (REL) dose of ARG would prove more effective in aiding performance by avoiding the potential side effects of high absolute (ABS) dosages (Forbes and Bell, 2011). Forbes and Bell (2011) revealed a significant increase in ARG plasma concentrations with relative dosages of 0.075 $g \cdot kg^{-1}$ and 0.15 $g \cdot kg^{-1}$ of body mass of ARG, though testing was performed under resting conditions. Therefore, the ergogenic potential of a REL versus ABS dose of ARG for exercise performance remains to be examined.

The aim of the present study was therefore to investigate the effects of relative versus acute dosages of L-arginine without any additional additives, on both aerobic and anaerobic performance in trained cyclists.

Materials and methods

Participants

Ethical approval was granted by the University of Central Lancashire Ethics Committee and in accordance to the Declaration of Helsinki. Participants were informed both verbally and in writing of the test procedures and written and informed consent was obtained. Eight healthy, trained male cyclists (mean ± *s*: age 21.00 ± 1.41 years, stature 176.13 ± 4.97 cm, mass 72.76 ± 3.40 kg) participated in the study. All had a minimum of 2 years competitive cycling experience and had previous experience of laboratory based cycle ergometry. Participants were required to abstain from alcohol and caffeine and from performing any strenuous activity for 24 hours prior to testing.

Instrumentation

Participants performed 1km and 16.1 km time trials (TT) on a cycle ergometer (SRM High Performance Ergometer, SRM, Jüllich, Germany). Saddle height, setback and handlebar position were adjusted to the participants personal preference and data were recorded at 5 s intervals during both trials. Following completion, mean power output (W_{mean}) and time to completion were calculated for each trial.

Capillary blood samples were collected from the left index finger to determine blood lactate (La) levels immediately post TT. Following cleansing using alcohol wipes the finger was punctured with a lancet (BD Microtainer, Dickinson and Co., Plymouth, UK). Blood lactate was determined using a portable lactate meter (Lactate Pro, Arkray, Kyoto, Japan) and measured in $mmol \cdot L^{-1}$. Supplementation used unflavoured L-arginine without any additional additives (L-arginine Powder, Now foods, USA) with 500 mL of water. Ambient laboratory conditions were temperature 17.67 ± 1.15 °C and barometric pressure 751.67 ± 4.89 mmHg.

Protocol

The study was a double blind, randomised design with participants acting as their own control. Participants

were required to attend three test sessions separated by a 7 day washout period. The sessions were defined as control (CON) where 500 mL of plain water was consumed, absolute (ABS) where 6 g ARG per 500 mL water was consumed based on the previous recommendation of Liu et al. (2009) and Bailey et al. (2010a). However, these recommendation do not take into account the influence of body mass, therefore a relative dose (REL) of 0.15 $g \cdot kg^{-1}$ body mass ARG per 500 mL was also investigated in line with the recommendations of Forbes and Bell (2011). Solutions were consumed 90 min prior to exercise. Previous research has shown that maximal concentration of plasma ARG is reached 90 min following the ingestion of 6 g of ARG (Bode-Böger, Bödger, Galland, Tsikas and Frolich, 1998). All solutions were flavoured with orange cordial to taste match and the order of testing was randomised.

During each session participants performed a 1 km and 16.1 km TT, separated by a 15 min passive recovery period. Prior to each TT participants performed a 10 min self-paced warm up on the SRM ergometer. Before each test a zero offset was performed to calibrate the SRM in accordance with the manufacturer's recommendations. During testing, participants received no temporal, verbal, or physiological feedback. Following completion of the each TT, La was recorded immediately. Participants completed a diet diary the week prior to the initial test session and were then instructed to maintain the same diet throughout the remainder of the study in order to minimise nutritional variations that may have influenced the results. All testing was conducted at the same time of day (± 2 hours) to reduce the effects of diurnal biological variation on exercise performance.

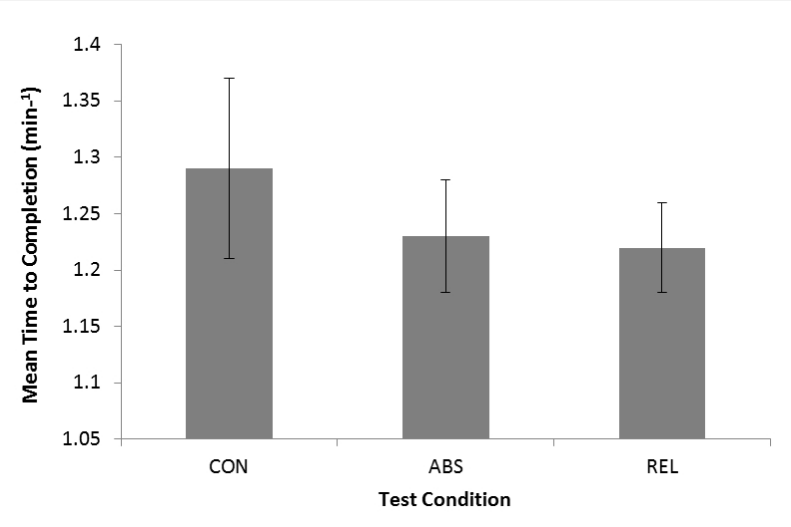

Figure 1. Mean time to completion for 1 km cycle time trials.

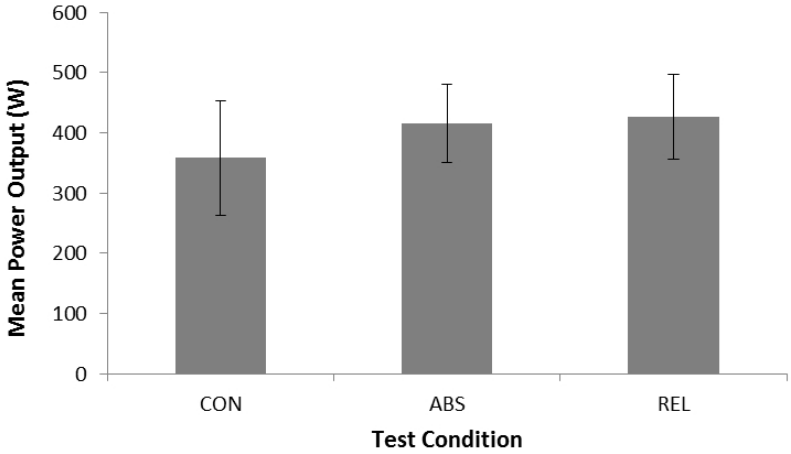

Figure 2. Mean power output during 1 km cycle time trials.

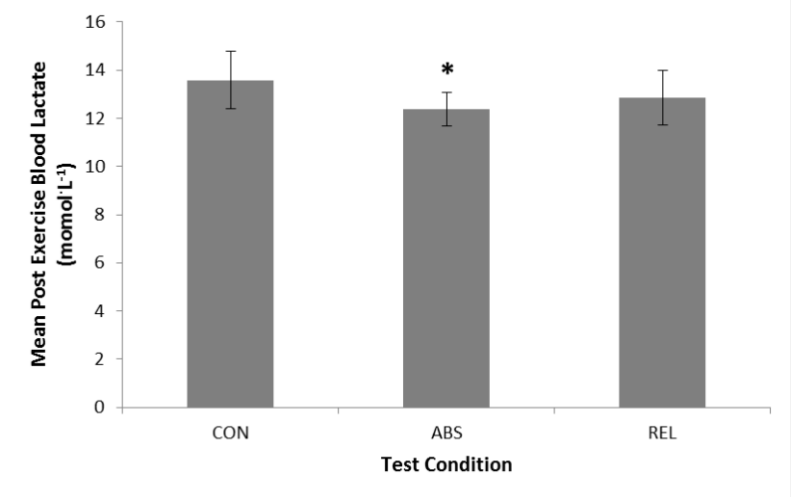

Figure 3. Mean post exercise blood lactate concentration following 1 km cycle time trials.
* Significantly different to control (CON) group.

Statistical analysis

Statistical analyses were performed using SPSS Statistical software (SPSS 20.0, SPSS Inc., Chicago). One-way repeated measures ANOVA's were used to test for differences between conditions. Where a significant main effect was found Bonferroni adjusted post-hoc pairwise comparisons were used to determine where the significant difference lay. An estimate of effect size (ES) was calculated using partial Eta squared (η^2). Based on the definition of effect size by Cohen (1988), >0.8 was considered large, ~0.5 as moderate and <0.2 as small. Statistical significance was accepted when $p \leq .05$. All values are reported as mean ± *s* unless otherwise stated. Statistical procedures were computed using SPSS v20 (IBM SPSS).

Results

1 km Time Trial

Statistical analyses revealed no significant main effect for time to completion of the 1 km TT by condition, $F(1.12, 7.83) = 4.64$; $p > .05$, $\eta^2 = .40$. However, there was a trend for time to decrease with supplementation. Mean time to complete the 1 km TT was 1.29 ± 0.08 min^{-1} for CON, 1.23 ± 0.05 min^{-1} for ABS and 1.22 ± 0.04 min^{-1} for REL, respectively (Figure 1).

No significant main effect was found for W_{mean} by test condition during the 1 km TT, $F(1.19, 8.32) = 4.68$; $p > .05$, $\eta^2 = .40$. Despite the lack of significance, there appeared to be a trend for W_{mean} to increase with supplementation over the CON trials (358.14 ± 94.73 W, 415.51 ± 65.36 W and 426.35 ± 70.86 for CON, ABS and REL, respectively) (Figure 2).

Statistical analyses revealed a significant main effect for La post 1 km TT, $F(2, 14) = 5.60$; $p = .02$, $\eta^2 = .45$. Post hoc analysis using Bonferroni correction revealed mean post 1 km TT La was significantly lower following ABS ($p = .04$) when compared to CON. No other significant differences were found. Mean post 1 km TT La values were 13.59 ± 1.21 mmol·L^{-1}, 12.38 ± 0.70 mmol·L^{-1} and 12.85 ± 1.14 mmol·L^{-1} for CON, ABS and REL, respectively (Figure 3).

16.1 km Time Trial

As with the 1 km TT, no significant main effect was reported for time to completion during the 16.1 km TT's, $F(2, 14) = 2.27$; $p = >.05$, $\eta^2 = .25$. However, there was again a trend for mean time to completion to decrease upon supplementation (27.27 ± 1.50 min^{-1}, 27.02 ± 1.58 min^{-1} and 26.52 ± 1.34 min^{-1} for CON, ABS and REL doses, respectively (Figure 4).

Data analysis revealed a significant main effect for W_{mean} during the 16.1 km TT's, $F(2, 14) = 10.05$; $p = .01$, $\eta^2 = .59$. Post hoc analysis found W_{mean} was significantly higher following a REL dose of ARG ($p = .01$) when compared to CON. Mean power output for the 16.1 km TT's were 196.19 ± 32.40 W, 206.31 ± 38.10 W and 215.81 ± 31.56 W for CON, ABS and REL, respectively (Figure 5).

Data analysis revealed a significant main effect for W_{mean} during the 16.1 km TT's, $F(2, 14) = 10.05$; $p = .01$, $\eta^2 = .59$. Post hoc analysis found W_{mean} was significantly higher following a REL dose of ARG ($p = .01$) when compared to CON. Mean power output for the 16.1 km TT's were 196.19 ± 32.40 W, 206.31 ± 38.10 W and 215.81 ± 31.56 W for CON, ABS and REL, respectively (Figure 5).

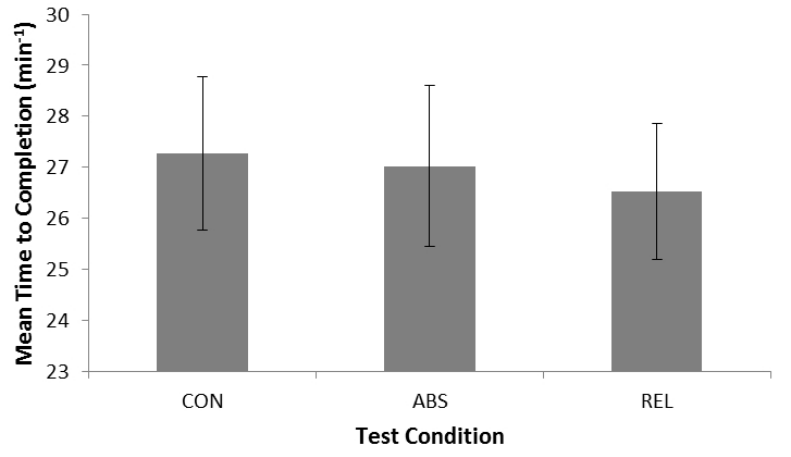

Figure 4. Mean time to completion for 16.1 km cycle time trials.

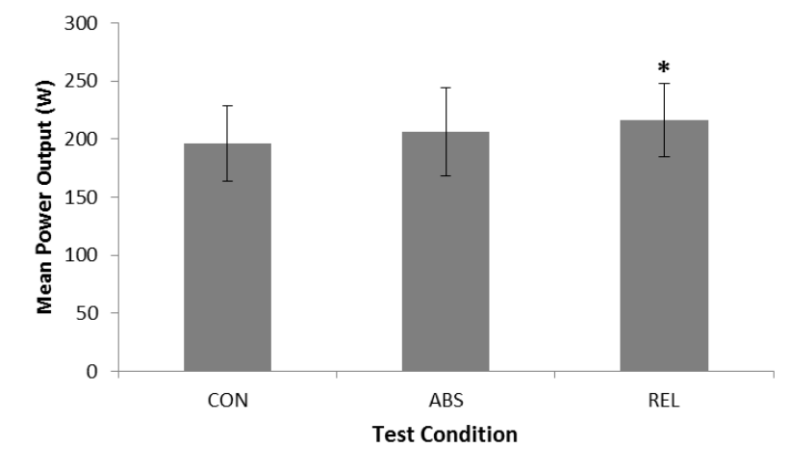

Figure 5. Mean power output during 16.1 km cycle time trials. * Significantly different to control (CON) group.

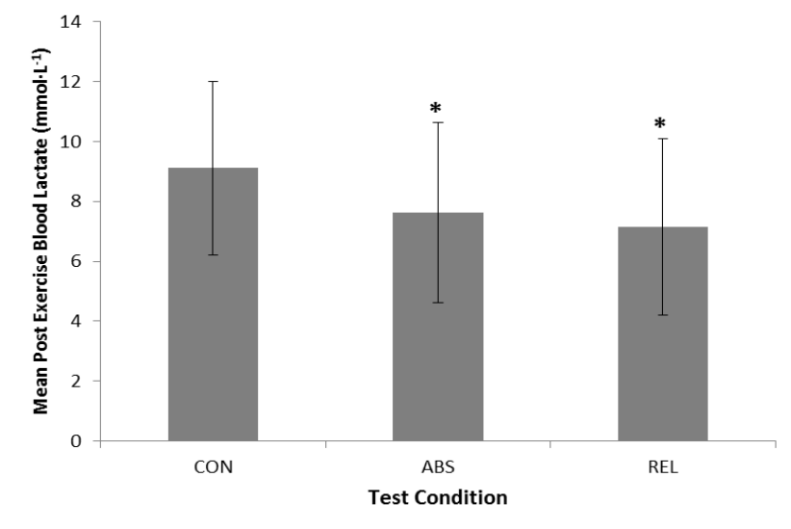

Figure 6. Mean post exercise blood lactate concentration following 16.1 km cycle time trials. * Significantly different to control (CON) group.

Discussion

The aims of the current investigation were to examine the effect of ARG supplementation without additives on 1 km and 16.1 km TT performance and to compare absolute and relative dosages of ARG.

The key findings of the study were that acute ARG supplementation significantly decreased 1 km and 16.1 km TT blood lactate values and increased mean power output during the 16.1 km TT in trained cyclists when compared to a control group. However, the differences in 1 km TT time to completion and mean power output between test conditions were non-significant, though only marginally (p = .06). When compared with CON values, mean 1 km TT time to completion showed a trend for improvement, as time to completion increased by 6.74 % and 7.87 % following ingestion of an ABS and a REL dose of ARG, respectively. Additionally, mean power output during 1 km TT also showed a trend for increasing following supplementation, though not to a level of significance, with power increasing 13.81 % and 16 % for ABS and REL, respectively when compared to CON.

Again, though not to a level of significance, time to completion decreased by 1.52 % and 2.13 % during the 16.1 km TT's following an ABS and REL dose of ARG, respectively. However, it is unclear why time to completion was not significant when mean power significantly increased during the 16.1 km TT. This may have been due to the relatively larger standard deviation of the time to completion results and therefore reducing the significance. In addition, pacing strategies may have resulted in higher power outputs at the start following supplementation, but dropping off more towards the end and therefore leading to the marginally non-significant differences in time to completion.

Though not significant, the improvements in TT performance following ARG ingestion elicited in the present study are considerably greater than the worthwhile change for 1 km (0.8-2.2 %) and 16.1 km road TT (0.9-2.4 %) in elite athletes proposed by Paton and Hopkins (2006). Therefore, these findings suggest that ARG supplementation with no additional compounds might have some potential to benefit athletic performance in events lasting 1-30 min in duration. However, the optimal dosage to achieve significance and the effects of ARG supplementation on performance in elite cyclists remain to be elucidated.

The mechanisms responsible for the observed changes in the present study are currently unclear. However, a possible explanation for the improvements observed in the 16.1 km TT could be an increase in NO production following the ingestion of ARG. However, the indices of NO production were not possible to measure in the current investigation and should thus be considered in future analyses. Nevertheless, improvements in time to task failure have been observed following ARG ingestion accompanied by increases in plasma NO_2^- elsewhere (Bailey et al. 2010a). It is accepted that plasma NO_2^- is an indicator of eNOS activity (Rassaf et al. 2007) and through its role as a physiological storage pool for NO production (Gladwin et al. 2006), provides an indication of high intensity exercise tolerance (Bailey et al. 2010a). Reductions in VO_2 slow component as a result of ARG supplementation (Bailey et al. 2010a) are thought to spare the utilisation of anaerobic reserves and the accumulation of fatigue inducing metabolites and reduce the ATP cost of muscle force production (Bailey et al. 2010b), subsequently improving exercise performance.

A possible mechanism responsible for the positive trends observed in the predominately anaerobic 1 km TT could be the role of ARG in creatine synthesis (Persky and Brazeau, 2001). It is well documented that increases in creatine production increase muscle phosphocreatine concentrations (Hultman et al. 1996), subsequently improving anaerobic power indices (Tarnopolsky and MacLennan, 2000).

Post 1 km and post 16.1 km La levels were all reduced following both ABS and REL dose of ARG compared to CON. However, the REL dose did not result in a significant decrease in La following the 1 km TT when compared with CON. To the researchers' knowledge, this is the first study to elicit reductions in La following supplementing with an oral form of ARG with no additional compounds. In addition, this study is the first to demonstrate a greater reduction following a REL dose compared to an ABS dose of ARG after a predominately aerobic exercise effort. Previous research has only found a reduction in La following a 3 g intravenous load of ARG (Schaefer et al. 2002) and 3 g oral intake of L-arginine-L-aspartate (Burtscher et al. 2005) in healthy subjects. Therefore, these results provide an insight into the potential La lowering abilities of a greater and more practical dose of ARG in trained individuals. The significant decreases in La during the 16.1 km TT may be indicative of greater glucose uptake during predominantly aerobic activity as proposed by McConnell and Kingwell (2006) and a reduced reliance on anaerobic metabolism (Mohr et al. 1996), whilst the non-significant results of blood lactate levels during the 1 km TT compared to the control group suggest that ARG supplementation may not necessarily be detrimental to predominantly high intensity anaerobic based exercise, where glycolysis is the primary energy provider and therefore the ability to tolerate production of high levels of lactate are a necessary.

Conclusions

In summary, the findings of the present study do not support the use of L-arginine supplementation for significantly improving cycling TT performance. However, the results did show a trend for reducing time to completion and small increases in mean power output. In addition, the results would appear to suggest that if athletes are to supplement with L-arginine, a relative dose may be more effective than absolute dosages. Further research is therefore warranted to determine the optimal dose to stimulate significant

changes and to further investigate the responses of blood lactate to nitric oxide production during exercise.

Practical applications

Based on the observations in the present study; it appears that ARG without any additional compounds in the dosages provided did not significantly enhance cycling performance. However, the results showed some trends that is could potentially provide a safe and effective method of improving cycling TT performance. Although the minimum dose of ARG supplementation required to improve performance significantly remains unknown, based on absolute doses, the results of the present study show an acute 6 g dose of ARG 90 min prior to performance can elicit small improvements. However, the present study also demonstrated slightly greater improvements in performance following a relative dose, as this better address the potential influence of body mass. Current research recommending a 6 g absolute dose may underestimate the ARG requirements of larger individuals when compared to an absolute dose. The present study would therefore indicate that relative dosing is more effective.

References

1. Álvares, T., Conte, C., Paschoalin, V., Silva, J., Meirelles Cde, M., Bhambhani, Y., and Gomes, P. (2012) Acute L-arginine supplementation increases muscle blood volume but not strength performance. Applied Physiology, Nutrition, and Metabolism, 37, 115-126.
2. Bailey, S., Winyard, P., Vanhatalo, A., Blackwell, J., DiMenna, F., Wilkerson, D., and Jones, A. (2010a) Acute L-arginine supplementation reduces the O_2 cost of moderate-intensity exercise and enhances high-intensity exercise tolerance. Journal of Applied Physiology, 109, 1394-1403.
3. Bailey, S., Fulford, J., Vanhatalo, A., Winyard, P., Blackwell, J., DiMenna, F., ... Jones, A. (2010b) Dietary nitrate supplementation enhances muscle contractile efficiency during knee-extensor exercise in humans. Journal of Applied Physiology, 109, 135-148.
4. Baydoun, A.R., Emery, P.W., Pearson, J.D., and Mann, G.E. (1990) Substrate-dependent regulation of intracellular amino acid concentrations in cultured bovine aortic endothelial cells. Biochemical and Biophysical Research Communications, 173, 940-948.
5. Benavides, G.A., Liang, Q., Dodson, M., Darley-Usmar, V., and Zhang, J. (2013) Inhibition of autophagy and glycolysis by nitric oxide during hypoxia-reoxygenation impairs cellular bioenergetics and promotes cell death in primary neurons. Free Radical Biology and Medicine, 65, 1215-1228.
6. Bescós, R., Sureda, A., Tur, J., and Pons, A. (2012) The effect of nitric-oxide-related supplements on human performance. The Journal of Sports Medicine, 42, 99-117.
7. Bode-Böger, S., Bödger, R., Galland, A., Tsikas, D., and Frolich, J. (1998) L-arginine-induced vasodilation in healthy humans: pharmacokinetic-pharmacodynamic relationship. Journal of Clinical Pharmacology, 46, 489-497.
8. Böger, R.H., Bode-Böger, S.M., Gerecke, U., and Frolich, J.C. (1994) Long-term administration of L-arginine, L-NAME, and the exogenous NO donor molsidomine modulate urinary nitrate and cyclic GMP excretion in rats – correlation with vascular reactivity. Cardiovascular Research, 28, 494-499.
9. Böger, R., and Bode-Böger, S. (2001) The clinical pharmacology of L-arginine. Annual Review of Pharmacology and Toxicology, 41, 79-99.
10. Burtscher, M., Brunner, F., Faulhaber, M., Hotter, B., and Likar, R. (2005) The prolonged intake of L-arginine-L-aspartate reduces blood lactate accumulation and oxygen consumption during submaximal exercise. Journal of Sport Science and Medicine, 4, 314-322.
11. Camic, C., Housh, T., Mielke, M., Zuniga, J., Hendrix, C., Johnson, G., ... House, D. (2010) The effects of 4 weeks of an arginine-based supplement on the gas exchange threshold and peak oxygen uptake. Applied Physiology, Nutrition and Metabolism, 35, 286-293.
12. Campbell, B., Roberts, M., Kerksick, C., Kerksick, C., Wilborn, C., Marcello, B., ... Kreider, R. (2006) Pharmacokinetics, safety and effects on exercise performance of L-arginine alpha-ketoglutarate in trained adult men. Nutrition, 22, 872-881.
13. Cohen, J. (1988). Statistical power analysis for the behavioral sciences 2nd Ed.). Hillsdale, NJ: Lawrence Erlbaum.
14. Flam, B.R., Eichar, D.C., and Solomonson, L.P. (2007) Endothelial nitric oxide production is tightly coupled to the citrulline-NO cycle. Nitric Oxide Biology and Chemistry, 17, 115-121.
15. Forbes, S., and Bell, G. (2011) The acute effects of a low and high dose of oral L-arginine supplementation in young active males at rest. Applied Physiology, Nutrition and Metabolism, 36, 405-411.
16. Gladwin, M., Raat, N., Shiva, S., Dezfulian, C., Hogg, N., Kim-Shapiro, D., and Patel, R. (2006) Nitrite as a vascular endocrine nitric oxide reservoir that contributes to hypoxic signalling, cytoprotection, and vasodilation. American Journal of Physiology Heart and Circulatory Physiology, 291, 2026-2035.
17. Hardy, T.A., and May, J.A. (2002) Coordinate regulation of L-arginine uptake and nitric oxide synthase activity in cultured endothelial cells. Free Radical Biology and Medicine, 32, 122-131.
18. Hishikawa, K., Nakaki, T., Suzuki, H., Katot, R., and Saruta, T. (1993) Role of L-arginine-nitric oxide pathway in hypertension. Journal of Hypertension, 11, 639-645.
19. Hultman, E., Soderlund, K., Timmons, J., Cederblad, G., and Greenhaff, P. (1996) Muscle creatine loading in men. Journal of Applied Physiology, 81, 232-237.
20. Jeacocke, N., and Burke, L. (2010) Methods to standardize dietary intake before performance testing. International Journal of Sport Nutrition and Exercise Metabolism, 20, 87-103.
21. Koppo, K., Taes, Y., Pottier, A., Boone, J., Bouckaert, J., and Derave, W. (2009) Dietary arginine supplementation speeds pulmonary VO_2 kinetics during cycle exercise. Medicine and Science in Sports and Exercise, 41, 1626-1632.
22. Liu, T., Wu, C., Chiang, C., Lo, Y., Tseng, H., and Chang, C. (2009) No effect of short-term arginine supplementation on nitric oxide production, metabolism and performance in intermittent exercise in athletes. The Journal of Nutritional Biochemistry, 20, 462-468.
23. McConell, G. (2007) Effects of L-arginine supplementation on exercise metabolism. Current Opinion in Clinical Nutrition and Metabolic Care, 10, 46-51.

24. McConell, G., and Kingwell, B. (2006) Does nitric oxide regulate skeletal muscle glucose uptake during exercise? Exercise and Sport Sciences Reviews, 34, 36-41.
25. Mohr, S., Stamler, J., and Brune, B. (1996) Posttranslational modification of glyceraldehyde-3-phosphate dehydrogenase by S-nitrosylation and subsequent NADH attachment. The Journal of Biological Chemistry, 271, 4209-4214.
26. Moncada, S., and Higgs, A. (1993) The L-arginine-nitric oxide pathway. The New England Journal of Medicine, 329, 2002-2012.
27. Morris, S. (2007) Arginine metabolism: boundaries of our knowledge. The Journal of Nutrition, 137, 16025-16095.
28. Olek, R., Ziemann, E., Grzywacz, T., Kujach, S., Luszczyk, M., Antosiewicz, J., and Laskowski, R. (2010) A single oral intake of arginine does not affect performance during repeated Wingate anaerobic test. Journal of Sports Medicine and Physical Fitness, 50, 52-56.
29. Paton, C., and Hopkins, W. (2006) Variation in performance of elite cyclists from race to race. European Journal of Sport Science, 6, 25-31.
30. Persky, A., and Brazeau, G. (2001) Clinical pharmacology of the dietary supplement creatine monohydrate. Pharmacology Reviews, 53, 161-176.
31. Rassaf, T., Lauer, T., Heiss, C., Balzer, J., Mangold, S., Leyendecker, T., ... Kelm, M. (2007) Nitric oxide synthase-derived plasma nitrite predicts exercise capacity. British Journal of Sports Medicine, 41, 669-673.
32. Schaefer, A., Piquard, F., Geny, B., Doutreleau, S., Lampert, E., Mettauer, B., and Lonsdorfer, J. (2002) L-arginine reduces exercise-induced increase in plasma lactate and ammonia. International Journal of Sports Medicine, 23, 403-407.
33. Schellong, S., Böger, R., Burchert, W., Bode-Böger, S., Galland, A., Frolich J., ... Alexander, K. (1997) Dose-related effect of intravenous L-arginine on muscular blood flow of the calf in patients with peripheral vascular disease: a $H_2{}^{15}O$ positron emissiontomography study. Clinical Science, 93, 159-165.
34. Tarnopolsky, M., and MacLennan, D. (2000) Creatine monohydrate supplementation enhances high-intensity exercise performance in males and females. International Journal of Sport Nutrition and Exercise Metabolism, 10, 452-463.
35. Wu, G., Bazer, F., Davis, T., Kim, S., Li, P., Rhoads, J., Yulong, Y. (2009) Arginine metabolism and nutrition in growth, health and disease. Amino Acids, 37, 153-168.
36. Wu, G., and Morris, S.M. Jr. (1998) arginine metabolism: nitric oxide and beyond. Biochemical Journal, 336, 1-17.